# Assessing the physiological costs of dominance in a highly social bird


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2016

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Todas as correções determinadas pelo júri, e só essas, foram efetuadas.
O Presidente do Júri,

Porto, _____/_____/_____

# Acknowledgments

I would like to express my deepest gratitude and debt of thanks to my supervisors and mentors Rita Covas, Claire Doutrelant and Sophie Lardy for the opportunity, advice, guidance, kindness and overwhelming enthusiasm that helped me to a great extent to accomplish this task. They allowed this study to be my own work, but steered me in the right direction whenever they thought I needed it, guiding me at every stage of my research. Their dedication and passion to the project was what made me follow the scientific journey of behaviour and to get where I am today.

For the laboratory expertise and undoubted physiology knowledge, I would like to thank to Benjamin Rey for the orientation, patience and lab assistance, as well for the sympathy and kind reception.

I would also like to thank Margaux Rat for developing the study of dominance in this species, for previously familiarize me and enlighten me on the subject and protocol, as well as teach me how to endure all the video analysis.

This work wouldn't be possible without the collaboration of wonderful people that helped at the field: Arnaud Tognetti, Caterina Funghi, Franck Theron, Kyle Loyld, Max Passerault, Myriam El Harouchy, Rita Fortuna and Thomas Pagnon.

I acknowledge De Beers Mining Corporation for providing access to the mesmerizing study field that is the Benfontein Game Reserve.

Additionally, I would also like to share a very special thanks to my dear master's colleague and friend Maria Teresa for the memorable friendship and restless company through all this journey and all the challenges and videos within.

I would also like to share my genuine gratefulness to my untiring and determined field partner André Ferreira, which creativity and knowledge are an inspiration to any researcher. I'm sincerely thankful for his patience, guidance and friendship.

Finally, I must express my very profound gratitude to my mother and friends (Ana A., Ana L., Ana P., Iris S., Joana B., Margarida S., Maria J., Nuno A., Paulo C. and Rita C.) for providing me with unfailing support and continuous encouragement through the process of researching, video analysis and writing, as well for the restless patience when I was miles away. This accomplishment would not have been possible without them.

# Abstract

The evolution and maintenance of social behaviour is a central question in behavioural ecology that requires an understanding of the possible trade-offs between the benefits and costs of group-living. Important costs of group living arise from an increase in competition among individuals, that may lead to both individual costs and conflicts of interests, which undermine group stability. Dominance hierarchies, and the associated evolution of signals of dominance (such as badges of status) often arise in these contexts to solve and prevent these conflicts by establishing an order of access to the different resources. Social dominance is expected to provide advantages in terms of access to resources, which is thought to translate in individual fitness benefits. However, acquiring and maintaining dominance status may also entail costs. For example, dominant individuals are likely to engage more frequently in aggressive behaviours to maintain their status or to obtain access to reproduction, or may invest energy and resources in signalling their status. These behavioural changes are likely to involve physiological challenges, through hormonal changes and higher metabolism, that may be related to the production of reactive oxygen species that, when not depleted by the individual antioxidant defences, can ultimately lead to oxidative stress. Oxidative stress is a known key mediator of life-history trade-offs, however the relationship between oxidative status and dominance remains seldom investigated. Here, I examine if there is a physiological cost, measured as oxidative stress, associated with dominance and associated signalling through a black throat patch (the “bib”) in a highly social and cooperative species of bird, the sociable weaver *Philetarius socius*. Oxidative status was assessed by measuring both the non-specific antioxidant capacity and the amount of reactive oxygen metabolites, while oxidative stress was assessed through the ratio of both markers. The findings of this work propose that dominance may be underpinned by oxidative stress in a sex-related manner, with dominant females showing higher levels of oxidants and oxidative stress than their subordinates. This suggests a physiological cost of dominance for females, while no costs were detected here for males, a difference that might be related to the sex-specific variation in oxidant defences found in this work. Males showed higher antioxidant levels than females, independently of the social rank. Signalling dominance through bib size was not related to any individual redox components, however bib size did seem to have a signalling function for females, with larger bibs found in dominant females. There was also a significant relation of patch size with body condition for both sexes. Therefore these findings show that social dominance

and associated signalling in cooperative species may entail a trade-off between advantages and hidden costs, and that costs can relate to oxidative status in a sex-specific manner.

Keywords: Social dominance, bib, badge of status, oxidative stress, ROMs, OXY, *Philetarius socius,* social rank, physiological costs, trade-offs.

# Resumo

A evolução e manutenção do comportamento social é uma questão central em ecologia comportamental que requer o estudo de possíveis compromissos evolutivos entre benefícios e custos da vida em grupo. Os custos mais determinantes da vida em grupo surgem da elevada competição entre indivíduos que pode levar a custos a nível individual, bem como a conflitos de interesses que ponham em causa a estabilidade do grupo. Hierarquias de dominância, e a associada evolução de sinais ("divisas de estatuto"), surgem neste contexto de forma a evitar este género de conflitos ao estabelecer uma ordem de acesso aos diferentes recursos. É expectável que a dominância proporcione vantagens em termos de acesso aos recursos, traduzindo-se em benefícios de aptidão para o indivíduo. No entanto, adquirir e manter um estatuto dominante também pode incluir custos. Por exemplo, indivíduos dominantes são suscetíveis de iniciar comportamentos agressivos ou de patrulha, de forma a manter o seu estatuto ou a obter acesso à reprodução, ou podem ter de investir mais energia e recursos em sinalizar o seu estatuto. Estas alterações comportamentais podem envolver alterações fisiológicas, como mudanças hormonais ou elevado metabolismo, relacionadas com a produção de espécies reativas de oxigénio que, quando não compensadas pelo sistema antioxidante do individuo, podem gerar stress oxidativo. Stress oxidativo é um reconhecido mediador de compromissos evolutivos e estratégias de vida, porém a sua relação com a dominância continua por ser investigada. Neste trabalho, pretendo investigar se existe um custo fisiológico, medido como stress oxidativo, associado com a dominância e sinalização através de um babete preto localizado sob o bico, numa espécie de passeriforme altamente social e cooperativa, o tecelão social *Philetarius socius.* O estado oxidativo foi avaliado através da capacidade antioxidante não-específica individual, como da quantidade de metabolitos de oxigénio reativos, enquanto o stress oxidativo foi calculado através do ratio entre os dois marcadores. Os resultados deste trabalho sugerem que a dominância pode estar relacionada com um aumento do stress oxidativo nesta espécie, mas de forma dependente do sexo, com fêmeas dominantes a exibir níveis mais elevados de oxidantes e stress oxidativo que as subordinadas. Estes resultados sugerem que existe um custo associado à dominância para as fêmeas, não detetado nos machos, que pode surgir da variação das defesas oxidantes entre sexos. Os machos exibem elevados níveis de antioxidantes, independentemente do seu estatuto social. O tamanho do babete não estava relacionado com nenhum componente oxidativo, mesmo tendo uma

função sinalizadora nas fêmeas, com babetes maiores encontrados nas fêmeas dominantes. Foi também encontrada uma relação significativa entre o tamanho do babete e a condição corporal em ambos os sexos. Assim sendo, estes resultados mostram que a dominância social e a sinalização a ela associada podem fazer parte de um compromisso evolutivo entre vantagens e custos em espécies cooperativas, e que estes custos podem estar relacionados com o stress oxidativo de forma diferente dependendo do sexo.

Palavras-chave: dominância social, babete, divisos de estatuto, stress oxidativo, ROMs, OXY, *Philetarius socius*, rank social, custos fisiológicos, compromissos evolutivos.

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# List of acronyms

AICc - Corrected Akaike information criterion.

AICs - Akaike information criterions.

LMMs - Linear mixed models.

LRT - Likelihood ratio tests.

OXY - Non-specific antioxidant capacity.

ROS - Reactive oxygen species.

ROMs - Reactive oxygen metabolites.

VIF - Variance inflation factor.

# Introduction

The evolution of social behaviour is an important question in ecology and evolution. Theories about the evolution and maintenance of group-living address a possible trade-off between benefits and costs, and the cost-benefit balance should be either neutral or positive in order to be evolutionary stable (Kappeler & van Schaik 2002). Multiple forms of benefits can be acquired depending on the ecological context and taxa, with many examples ranging from cooperative acquisitions of food, to information transfer or reduced threat of predation due to dilution effect (Van Schaik 1983; Wrangham 1980). However, costs are also a determinant component of the evolutionary trade-off affecting sociality. For example, group living is associated with increased risk of disease transmission (Altizer *et al.* 2003) and significant costs of group living arise from an increase in competition among group members and by having unequal positions in the group.

Conflicts among group members, on whether to compete or share resources, have profound fitness implications by undermining group stability (Poisbleau *et al.* 2006) and species have developed a large assortment of behavioural strategies to mitigate such negative aspect of group living. One very common strategy is the definition of an order of access to the available resources upon the establishment of dominance hierarchies and associated evolution of signals of dominance, named badges of status (Senar 2006). Badge of status communicate social status by providing information about individual fighting ability, aggressiveness, motivation and other context-dependent factors, through variation in badge size or intensity, enabling rival assessment without confront (Senar 1999; Senar 2006). Dominance hierarchies and badges of status can be found in many taxa, from insects to primates (Hughes 1992; Laubach *et al*. 2013; Röseler *et al.* 1984; Gerald 2001; Thompson & Moore 1991).

The relative position of an individual within its social environment may have considerable impacts on its fitness. Individuals attaining a higher dominance status usually are the ones that obtain higher access to or monopolize high-quality resources (e.g., food, territories or breeding opportunities), which may have important positive short- and long-term fitness consequences, influencing not only survival (White 2007), but also dispersal (Chiarati *et al.* 2011) and reproductive success (Côté & Festa-Bianchet 2001; Nelson-flower & Ridley 2015). For example, high-rank primates appear to have greater foraging efficiency (e.g. grey-cheeked mangabeys *Lophocebus albigena,* Chancellor & Isbell

2009) and energy intake rates (e.g. white-faced capuchin *Cebus capucinus*, Vogel 2005) than low-rank ones, while in acorn woodpeckers *Melanerpes formicivorus* societies, the large and dominant brood mates experience greater survival and were more successful at gaining reproductive opportunities (Koenig *et al.* 2011). As a consequence of these clear benefits of being dominant, many early studies on social status and sociality focused on the benefits of being dominant as opposed to the costs of being subordinate. Low social status was viewed as stressful and costly, due to increased received aggression rate, poor nutrition and reproductive suppression (Creel *et al*. 2013, and references therein). However over the last decade, studies have been showing that attaining a higher dominance rank can also be costly and may have negative impacts on individuals (Creel *et al*. 2013).

Indeed, acquisition and maintenance of a higher dominance status could entail important costs, mostly when dominance ranks are unstable. Such costs may arise from frequently engaging in energetically and potentially injurious costing fights or aggressive behaviours to maintain their status through "policing", or to obtain access to reproduction (Clutton-Brock *et al.* 2006; Ang & Manica 2010), challenges rarely faced by subordinates (Creel *et al.* 2013; Creel 2001; Sapolsky 2005). Additionally, the higher rates of reproduction, characteristic of dominant individuals, may impose great energetic and physiological costs linked with demanding activities such as gamete production, mate acquisition and parental investment (Alonso-Alvarez *et al.* 2004; Cram *et al.* 2014; van de Crommenacker *et al.* 2011). Finally, dominant individuals may additionally invest more energy in signalling their status (Senar 1999).

In birds, dominance status is often signalled through plumage colourations functioning as badges of status (Senar 2006). Although these signals decrease the costs associated with aggression and escalated fights, they may bring other costs. For a signalling system to be evolutionarily stable, honesty must be maintained and cheating must be controlled (Maynard-Smith & Harper 2003). Badges of status are often melanic, and the honesty of this type of signal is often insured by physiological costs linked with the production and maintenance of this colouration, display (in terms of energy, resources and time; Roulin 2015), the increased production of androgens (Bókony *et al.* 2008) that may interfere with the immune system (''immunocompetence handicap"; Poiani *et al.* 2000) and with pleiotropy (e.g. shared genetic and biochemical pathways between melanogenesis and body condition; Roulin 2015). Hence, individuals with larger colourations should suffer higher costs. By contrast under the social cost hypothesis, badge of status would have costs ensured by social challenges and assessment of superiority of individuals displaying similar badges size (Maynard-Smith *et al.* 1988, Maynard-Smith & Harper,

2003). Under this hypothesis, intermediate badge size animals, which are more common, should fight more, which may lead to stress and/or injury and, ultimately, to higher physiological costs than individuals displaying large bibs (Acker *et al.* 2015). These costs associated with dominance and signalling, as well as the mechanisms mediating these costs, remain seldom investigated (Beaulieu *et al.* 2014; Cram *et al.*, 2014; Creel, 2001; Georgiev *et al.* 2015).

Oxidative stress refers to the detrimental accumulation of oxidative damage in cells and tissues, an accumulation greatly caused by an imbalance between reactive oxygen species (ROS) and organism antioxidant defences (Pamplona & Costantini 2011; Finkel & Holbrook 2000). ROS, free radical and non-radical oxidants, are known by-products of the aerobic metabolism through the mitochondrial respiration (Finkel & Holbrook 2000; Nemoto et al. 2000). ROS are highly reactive molecules capable of triggering a cascade of oxidative events that are important for the cell function, through signalling and immune function (Nemoto et al. 2000; Babior et al. 1973). However, when not sustained by the body enzymatic and non-enzymatic, endogenous and exogenous antioxidant defences components of the antioxidant machinery (reviewed in Monaghan *et al.,* 2009), this cascade has an extensive deleterious effect on key biological molecules such as lipids, proteins and DNA, damages known to be associated with pathology and senescence (Finkel and Holbrook, 2000). This disequilibrium between an increased production of ROS overpowering depleted defences has been proposed as a physiological mechanism underlying life history trade-offs (Monaghan *et al*. 2009; Speakman *et al*. 2015). Although there is no direct link between energy production and ROS production (see Salin *et al., 2015;* Speakman *et al*., 2015), ROS levels may be associated with metabolic activity and the allocation of energy between crucial body functions and oxidative defences (Finkel & Holbrook 2000), a relationship ultimately linked to metabolic investment in growth and reproduction (Ellison 2003; Arnott *et al.* 2006). Moreover, ROS production was found to be closely linked with age (Wegmann *et al.* 2015), sex (Isaksson 2013; Beaulieu *et al.* 2014), body condition (Finkel & Holbrook 2000), reproductive investment *(Georgiev et al.* 2015), aggression and territorial defence (Rammal *et al.* 2010), signalling (Vitousek *et al.* 2013) and social status (Beaulieu *et al.* 2014).

The physiological challenges faced by dominant individuals may involve higher energy expenditure and maintaining higher levels of stress hormones and testosterone, which are both associated with the aggression involved in warding off competitors (Wingfield *et al.* 1987) and in signal production and maintenance (Bókony *et al.* 2008). The production of such stress, aggression and signalling related hormones are expected to be linked to an increase in the production of ROS (Constantini & Hackney 2013; Alonso-

Alvarez *et al.* 2007). For instance, in male territorial albino mice *Mus musculus* aggressiveness, which is usually linked to dominance, was highly linked with ROS generation in peripheral granulocytes, and thus, with oxidative status (Rammal *et al.* 2010). On the other hand, in mandrills *Mandrillus sphinx* ROS was related with status in a sex-specific manner with only dominant females showing a higher levels of oxidative stress (Beaulieu *et al.* 2014), while in the seychelles warblers *Acrocephalus sechellensis* the dominant breeding males were the ones showing higher levels of circulating oxidized molecules (van de Crommenacker *et al.* 2011). Similarly, antioxidant defences may also be associated with individual status, as found in sparrow weavers *Plocepasser mahali* where dominant females showed significant reductions of antioxidant defences throughout the breeding season (Cram *et al.,* 2014). The latter examples highlight the differential burden and risk that aggressive or dominant individuals may undertake and influence individual oxidative status, however counter examples exist (Georgiev *et al.* 2015; Nelson-flower & Ridley 2015) and the relation between dominance and oxidative status is still unclear.

Furthermore, from the knowledge that the cumulative exposure to oxidative stress is negatively linked to individual life-history components, such as future reproduction and longevity (Costantini & Omo 2015; Geiger *et al.* 2012), measuring the components regarding individual oxidative status, assessing both the antioxidant machinery and oxidized molecules (Costantini & Verhulst 2009), seems a promising tool to decipher the physiological costs underlying dominance and, ultimately, underlying the life-history trade-offs regarding the social environment.

Here, I investigate if there is a physiological cost, measured as oxidative stress, associated with individual dominance rank in a highly social, colonial and cooperatively breeding species of bird, the sociable weaver *Philetarius socius*. These birds live in a non-egalitarian society, with colonies structured by strongly ordered and stable hierarchies, and males being dominant over females. Dominance hierarchies are influenced by relatedness to dominant colony members and individuals' aggressiveness, and are signalled by a melanin-based bib, possibly working as a badge of status *(Rat et al.* 2015). Additionally, dominant individuals were found to obtain privileged access to food (Rat *et al.* 2015) and reproduction (Rat *et al. submitted*). Aggressive interactions between sociable weavers rarely involve considerable injury or death (personal observation). Nevertheless, in this species dominance status was found to be related with bib size, which was also found to be linked with survival (Acker *et al.* 2015). As fights rarely result in injury, this survival differences may suggest a hidden, but determinant, physiological cost, such as oxidative stress, involving social interactions and the

physiological and/ or social costs of the melanin-based coloration (Roulin 2015; Vitousek *et al.* 2013).

Therefore, I expect a close link between individual variation in oxidative status, social rank and the melanic black bib. To investigate this link I assessed individual's dominance status by scoring interactions over a food resource, and test if dominance is related with two relevant metrics, which involve: 1) the oxidant component, by measuring the concentration of reactive oxygen metabolites (ROMs) in plasma, a proxy of oxidative damage; and 2) the antioxidant component by assessing non-specific antioxidant capacity (OXY) of plasma. It is crucial to measure both components to avoid misinterpretation since it can be misleading to assess only one side of the oxidative balance, either by assessing only oxidants production, since higher levels can be mitigated by efficient defences, or by only assessing antioxidant defences that may not be sufficient to deal with the levels of oxidants present (Monaghan *et al.* 2009). Therefore, the ratio between both was additionally calculated as a measure of oxidative damage. I also tested if the costs of badge of status production can be reflected by these two markers, and thus, on oxidative balance.

If dominance is costly, I expect higher values of oxidative stress in higher rank individuals, characterized also by larger bibs, both linked with a higher oxidative damage (represented in higher ROMs levels) and/or weaker antioxidant protection (represented by lower values of OXY). However, subordinates are also expected to pay higher survival costs, associated with aggression and poor nutrition, so a similar prediction can be made that they will also experience great oxidative stress levels. By contrast, if costs of sociality are higher for middle rank individuals, who were found to engage more frequently in status assessment fights (Acker *et al.* 2015), higher levels of oxidative stress would be expected for medium-ranked birds. Finally, I expect sex-specific differences in oxidative status, with higher values of ROMs and depleted oxidant defences in females, as a result of sex-related differences in dominance status and the different reproductive strategies pursued by males and females, arising from differential breeding effort and workload (Monaghan *et al.* 2009; Wegmann *et al.* 2015; Costantini *et al.* 2008; Ellison 2003).

# Methods

## Study species and field site

The sociable weaver is a colonial and facultative cooperatively breeding passerine that inhabits the semi-arid savannahs of the southern Kalahari and Namib regions of Namibia and of South Africa's Northern Cape Province. This weaver species lives in large colonial nests, used for both roosting and breeding (Maclean., 1973c). These structures are cooperatively build and made mainly of *Stipagrostis* grasses and assembled on a variety of sturdy structures, with Acacia trees being the most common. Sociable weavers inhabit a highly fluctuating environment characterized by an unpredictable rainfall, both in timing and quantity. Rainfall is the main determinant of food availability through its effects on the abundance of seeds and insects on which these weavers feed (Maclean., 1973c) and, consequently, is determinant for breeding activity and success (Covas *et al.* 2008). Sociable weavers may breed in pairs or cooperative groups with 1-5 helpers (Covas *et al.* 2008*)*. As in most cooperative systems (e.g. Lardy *et al.* 2015; Nelson-Flower *et al.*, 2011), sociable weavers' access to reproduction is skewed and associated with dominance (Rat. *et al.,* submitted) and age (Covas *et al.* 2006). Helpers are usually younger birds that postpone reproduction in the first years of life and help the parents raise their siblings. Sociable weavers form ordered hierarchies and have a melanin-based black throat bib, known as bib (Fig. 2), that was found to work as a badge-of-status with bib size reflecting dominance status (Rat *et al.* 2015). These weavers are sexually monomorphic and hence both sexes have black bibs, but males are usually dominant over females.

Field work was conducted from August to early September 2015 at Benfontein Game Farm (28° 52′ S, 24° 50′ E) near Kimberley in the Northern Cape Province, South Africa, an area of open savannah encompassing ca. 30 sociable weaver colonies varying in size (from 5 to 130 individuals). This investigation was part of a long-term research project into the cooperative behaviour and population dynamics of this species. To this end, every year 13 colonies are captured at the study site using mist nests. When captured, all individuals are ringed with both a numbered metal ring and a unique colour combination, allowing for individual identification at recapture and video recordings. During captures, blood samples are taken to genetically determine sex, parentage and relatedness (Griffiths *et al.* 1998). Sex was determined by amplification of chromo-helicase-DNA-binding genes located on the W and Z sex chromosomes using the P2 and P8 universal primers (Griffiths *et al.* 1998). The analysis were conducted at the University of Sheffield (UK). All individuals are weighed (to the nearest 0.01g), and wing

and tarsus length measured (to the nearest 0.5 and 0.01mm, respectively). In addition, I photographed the bib of each individual bird to allow measuring bib size, by using an established protocol (Rat *et al*., 2015; see below). For this study I used 5 colonies captured by the team (including myself). Colony size was estimated by counting the number of birds caught and adding the number of birds that were seen escaping by avoiding the nets (capture success varies between 85-100%, R. Covas unpublished data).

These captures at the beginning and end of the breeding season and regular visits to the nests during the breeding season to ring the nestlings allows either to accurately assess the age of the individuals marked as nestlings, or to estimate a minimum age during the captures.

## Dominance and behaviour analysis

Dominance hierarchies within five colonies were determined by myself using an already established protocol based on the behavioural analysis and scoring of agonistic interactions between individuals when feeding at an artificial food source (a mixture of bird seeds fed *ad libitum*) in a red-brown plate placed underneath each colony (Rat *et al.*, 2015, Fig. 1). Behavioural observations were performed using a video camera (Sony Handycam HD) on a tripod 2-3 m close to the feeder, allowing to record all interactions up to 1 m around the feeder. Two hours of observations were conducted every day, for 11.6 ± 0.730 SD days per colony, with the recording process starting on the 28 of August 2015. These observations were done between 9:00-10:00 and repeated in the afternoon between 14:00-15:00, for a total of 213 hours (each colony with a total of 42.6 ± 2.794 SD hours). The feeder was removed between recordings to increase competition and, thus the number of interactions observed (Rat *et al.* 2015).

I scored both the type and direction of agonistic interactions observed (aggressions, displacements, threats and avoidances, see Table 1 for a summarized description). For each interaction the identity of the birds was also scored to assess the “winner” and the “looser”. In this species, the initiator is always the one determining the encounter outcome and considered as winner in all types of interactions, except for avoidance that was the opposite (who avoided was considered as looser). To define a bird as resident at the colony, and to include possible immigrants and avoid prospecting ones (i.e.. individuals that don’t roost at the colony), I defined a line of criteria for colony attribution: I considered a bird as resident at the colony if it was captured at that colony or, when not captured, if it appeared at least in 3 consecutive days at that colony (both criteria

matching the number of interactions and the colony which the individual was last captured). For individuals captured in more than one colony or that were recorded over more than 3 days in more than one colony, I considered the colony where they appeared for more days (again matching the number of interactions obtained).

I recorded a total of 17814 agonistic interactions in the five studied colonies (3568.2 ± 1427.355 SD interactions per colony), from which dominance scores were calculated and obtained for 167 adult individuals (64 females, 90 males and 13 with unknown sex). These interactions were as follows: 4045 aggressions, 6739 displacements, 1463 threats and 5567 avoidances. Using the direction of the interactions, David's score (David 1987) was used to determine each individual dominance score (for each colony). David's score appears to be the most appropriate method to assess individual rank, through individual overall success, since it's based on the unweighted and weighted sum of individual proportions of wins by taking into account the relative strength of its opponent (Gammell & de Vries 2003). This score is then calculated by the weighted sum of individual proportion of wins (weighted by the wins of the adversaries) minus the weighted proportion of losses (weighted by the losses of its adversaries), using the "steepness" R package (De Vries *et al*. 2006). Following Gammell and de Vries (2003) procedures, social dominance scores were standardized using: $\frac{DSi-DSmin}{DSmax-DSmin}$ ; here ί represents the individual score within the colony, $DS_{min}$ *and* $DS_{max}$ as the lowest and highest scores obtained in that colony. The final values were ranged from 0 to 1 (from the most subordinate to the most dominant individual) and allowed the comparison of scores between colonies of different sizes.

**Fig. 1** Sociable weavers interacting at the feeder

**Table 1.** Description of scored agonistic interactions at the feeder used to calculate individual David's score

| Type of Agonistic Interaction | Description |
|---|---|
| Aggression | Individual A physically attacks individual B by pecking, kicking or wing flapping. |
| Displacement | Individual A moves directly towards a point of intersection with individual B until B moves away. Also includes chasing. |
| Threat | Individual A intimidates individual B using the beak, inflating the head and adapting a conspicuous position until B moves away, without involving physical contact. |
| Avoidance | Individual B moves away from individual A, evading A without A moving directly towards to B. |

## Bib size estimate

To estimate the black bib size I followed a protocol similar to Rat *et al*. (2015), with each individual photographed three times using a Nikon COOLPIX AW120. However, for this study each individual was photographed inside of box specially built for this purpose (Fig. 2), where each individual was hold and photographed by myself against a scaled neutral white background in a standardized position. Photographs were taken perpendicular to the head and the beak kept towards the optical lens. Between photos the feathers were always repositioned. Incomplete bibs of nestlings and juveniles were not included to avoid the variation in plumage maturation related to age. I measured the bib using Photoshop CS6 (Version 13.0.1.3) by selecting the bib black pixels and calculating the size in $cm^2$ through the background scale (by selecting the pixels contained in 1 $cm^2$ of the paper, a selection made for each photo and then averaged for each session). The final value that was used in my analyses was the mean bib size estimate of the three photos. Bib size was normally distributed with estimates ranging from 1.115 to 1.920 $cm^2$, and with a mean±SD = 1.425±0.168 $cm^2$, a range quite similar to the one obtained before (1.40±0.22 SD $cm^2$, Acker *et. al* 2015). Repeatability between measurements of photos of the same individual was based on an intra-class correlation coefficient that relies on the use of mixed-effects models for extracting the partition of variance's components and is calculate in the R package "rptR" (Nakagawa *et al.* 2007). The within-individual repeatability of bib size measures was found to be reasonably repeatable 0.782±0.032 SE ($P<0.001$, N= 298).

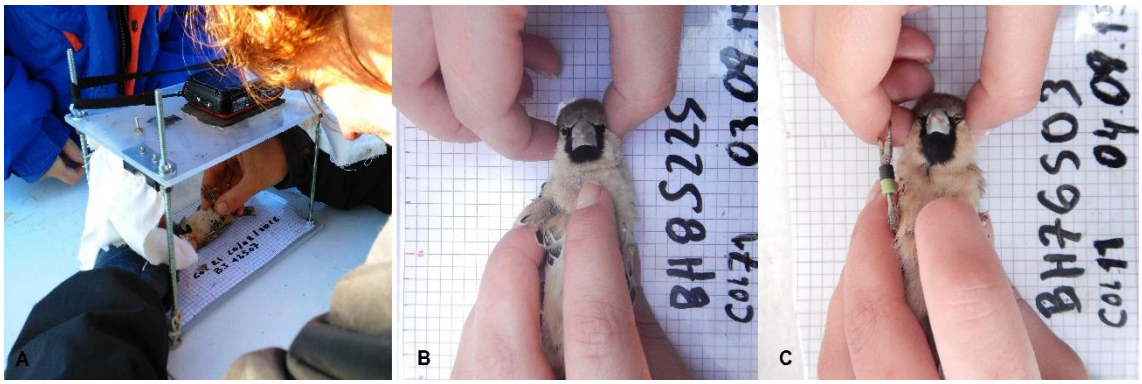


**Figure 2**: A) Box specially built for the standardization of the black bib photograph procedures at the field. Examples of two photographs of the sociable weaver's black bib used for bib size analysis: B) Individual displaying a small bib (1.186 $cm^2$). C) Individual displaying a larger bib (1.733 $cm^2$).

## Oxidative stress assays

A blood sample (c. 75 µl) was taken from each bird from the brachial vein using a sterile needle and a heparinized capillary tube. The time of sampling in the day, as a proxy between capture and the completion of blood sampling and almost immediate release, was recorded. Blood samples were centrifuged in the field at 6000 rotations for 2 minutes (Sprout minicentrifuge, Fisher Scientific, UK) and directly after placed in a cooler box until frozen and stored in the field station. The plasma was then frozen at -20 ºC at the field station, shipped back to Europe in dry ice and stored at -80°C until further analysis.

I performed the oxidative status analysis at the CNRS "Biométrie et Biologie Evolutive" laboratory, University of Lyon, Villeurbanne, France. Individual's oxidative profile was assessed by measuring concomitantly the i) circulating oxidative damages measured by the amount of reactive oxygen metabolites, ROMs) and ii) non-specific antioxidant capacity of plasma. Both components were quantified using commercial determination kits, the d-ROM and OXY-adsorbent tests respectively (Diacron International, Grosseto, Italy), following the protocols developed by Costantini & Dell'Omo (2006), with few minor modifications.

ROMs are an indirect measure of primarily hydroperoxides (ROOH), free radical ROS derivatives that are produced in the oxidative cascade early phases, providing a reliable marker of oxidative damage (Costantini & Dell'Omo 2006; Alberti *et al*. 2000). This group of ROMs is a suitable measure of oxidative status since, despite their oxidant power, these reactive species are relatively more stable (when kept at -80°C) and easy to detect compared to other relevant metrics (e.g. marker lipid peroxidation, Isaksson, 2013). Although there are few concerns in the literature that ROMs assays may be insensible

to environmental changes (Lindschinger *et al.* 2004) or affected by ceruloplasmin (Kilk *et al.* 2014), this marker has been widely recognized as a useful and highly repeatable (Costantini & Dell'Omo, 2006, Constantini, 2016) indirect measure of oxidative stress in several ecological studies (Isaksson 2013; Herborn *et al*. 2011; Beaulieu *et al.* 2014; Van de Crommenacker *et al.* 2011; Costantini & Dell'Omo 2015). The plasma (c. 20 µl) was first diluted with a 202 µl solution containing 200 µl of an acetic acid/sodium (pH 4.8) acetate buffer and a 2 µl of a chromogenic mixture calibrator (*N,N*-diethyl-*p*-phenylenediamine). The same volume was used for the blank. In the d-ROM test, the presence of an acidic buffer causes the release of metal ions from proteins of the plasma that will cleave the metabolites in alkoxyl and peroxyl radicals. Such radicals, in turn, will react and oxidize the alkyl-substituted aromatic amine soluble in the chromogenic mixture, resulting in pink-coloured complex photometrically quantified. The intensity of this coloured complex is directly proportional to the concentration of the sample' ROMs. After incubation (90 min), the absorbance was read with a microplate reader at 490 nm (SAFAS MP96-UV, Monaco) and compared with the reference curve using a standard, using: $\frac{ABSs}{ABSc} \times C$ , where $ABS_S$ and $ABS_c$ refers to both the absorbance of the sample and the calibrator, and *C* the concentration of the calibrator added. The final concentrations are expressed in mg H2O2/dl. The higher the concentration of ROMs, the higher the oxidative damage detected in the plasma. For the replicates, I included all captured colonies and all the samples from 2015 used for parallel studies, identifying each sampling date for the same individual as a distinct sample (to avoid the confounding effect of the individual state at each sampling time). Both the repeatability within (N=722) and between plaques (N=32) was found to be quite high with 0.920 ± 0.008 SE and 0.849 ± 0.084 SE, respectively (P<0.001).

The plasma antioxidant capacity measured by the OXY-Adsorbent test is assessed through the ability of the plasma antioxidant barrier to cope with the massive oxidant action of hypochlorous acid (HClO). This test includes a large range of antioxidant compounds of exogenous origin (e.g. tocopherols, carotenoids and flavonoids) or endogenously synthetized (e.g. bilirubin, cholesterol and glutathione). This test seemed the most appropriate for this type of analysis since it requires small amounts of plasma, reducing the impact in the species, and allows portraying the antioxidant properties of many antioxidant compounds (and not a single antioxidant). Additionally the contribution of the uric acid to the measured antioxidant capacity has been proved to be low (Costantini 2010; Récapet *et al.* 2016), therefore avoiding the confounding effect of accidental oxidation activity, since uric acid has both antioxidant and oxidant properties. Finally, both d-ROM and OXY-Adsorbent tests could be prepared and performed

simultaneously. All important criteria for markers choice, as reviewed by Horak and Cohen (2010). For the OXY-Adsorbent test, first 5 µl of plasma was diluted in 490 µl of distilled water and 10 µl of calibrator (serum MC030). The same volume was used for the blank. Then 200 µl of HClO solution was incubated (10 min) with the 5 µl of the diluted plasma, then adding 10 µl of the chromogen mixture (*N,N*-diethyl-*p*-phenylenediamine). This solution clearly exceeds the adsorption capacity of the antioxidant barrier so that the residual non-processed HClO oxides the alkyl-substituted aromatic amine chromogen added, transforming it in a pink-derivate which absorbance was then read with the microplate reader and finally compared with the reference curve of the calibrator using: $\frac{ABSb-ABSs}{ABSb-ABSc} \times C$ , where $ABS_b$ refers to the absorbance of the blank. OXY is presented as mM of acid neutralized, and the higher the OXY levels, the better the defences. For all the replicates (also including all colonies and all the samplings), the repeatability within (N=924) and between plates (N=116) was quite different, with the within plate replicates showing again a quite high repeatability 0.905 ± 0.008 SE, however contrasting with a moderately low repeatability found between plates 0.407 ± 0.11 SE (P<0.001).

Finally, since oxidative stress can only be assessed by the parallel of the two measures, an index was calculated through the ratio of ROMs to OXY (x1000) (Costantini *et al*. 2006b).

## Statistical analysis

To test whether individual oxidative status was predicted by individual dominance rank, I performed three separate linear mixed models (LMMs) with ROMs levels, OXY levels or ratio as dependent variables and dominance score as main effect.

Since other confounding variables could also influence individual oxidative status I also included the following: sex, minimum age in years (range of 1-8 years), time of sampling (ranging from 7h56 to 11h41) as in my exploratory analysis this was found to have a highly significant effect on both markers (LMM: p<0.01), colony size as this was also reported to have an effect on oxidative status in other species (e.g. Cram, Blount, & Young, 2015; Lardy *et al*., 2016) and there may be stronger competition in larger groups, mass (23.45-30.07 g) and tarsus length (22.24-24.68 mm) as a measure of body condition. Body condition was also assessed through the residuals of the regression mass-tarsus and showed similar results when replacing tarsus with mass. The most biologically relevant interactions terms: dominance*sex, dominance*age and dominance*sex*age were tested. Colony identity was used as random effect in all

models to account for a possible correlation of behaviour and physiology within colonies and avoid pseudoreplication.

To test if the production and maintenance of the melanin-based black bib has an oxidative cost, I conducted a similar analysis to the one above with three separated models with the oxidative status components but with bib size as the main effect instead of dominance score. Acker *et al*., (2015) found a U-shaped relationship between survival and sociable weavers' bib size, therefore I also tested for a possible quadratic relation with the markers by including both linear and quadratic components in the model.

To address the possible correlates that may also affect bib size variation I also performed a linear model with bib as dependent variable, and the interaction between sex and age, body condition and colony size as independent variables.

Finally, to confirm the findings of a previous study on the signalling function of the black bib (Rat *et al.* 2015) and assess the relationship between bib size and dominance, as well as potential correlates of dominance (age, sex and body condition), I also performed a linear mixed model with the dominance score as dependent variable and, again, controlling for sex, age and body condition as explanatory variables. I also tested for the most relevant interactions terms: age*sex, bib size*sex, bib size*age and bib size*sex*age, as well for a possible quadratic relation with dominance as above.

Previous studies have revealed a positive correlation between oxidants production and antioxidant defences as a possible and compensatory mobilization of defences against the free radicals (Costantini *et al.* 2010). Therefore, I also tested if individual ROMs levels were correlated with OXY defences. For this analysis I performed a Pearson's correlations, using the R package "Hmisc" (Harrell & Dupont 2016).

In order to avoid potential multicollinearity problems, Spearman rank correlations for non-normally distributed variables, and Pearson's correlation for normal distributed variables, were used to test possible correlations between the explanatory variables (dominance score, sex, minimum age, tarsus, mass, bib size). Only one set of variables was found to be strongly correlated, both sex and dominance score ($r=0.62$, $N=154$, $P<0.01$; for the remaining: $r<0.46$). To assess the importance of this correlation on variance inflation of my models and avoid possible results distortion, I calculated the variance inflation factor (VIF) using the "AED" R package (Zuur *et al.* 2009). As recommended by the authors, all values were <5 and, therefore, I included the confounding factor of sex in all of my models following the same procedures described before.

For all models, a linear mixed models approach was performed using the R package "lme4" (Bates, 2009). To verify the normality assumptions of linear mixed models, residuals were analysed using QQ plots, fitted versus residuals plots and histograms. Regarding model selection techniques, there is a current debate in the literature about the advantages and pitfalls of the different methods (Stephens *et al*. 2007), more specifically on whether model selection should be conducted through classical null hypothesis significant testing (e.g., stepwise model selection, Guthery, Brennan, Peterson, & Lusk, 2005), or model selection using corrected Akaike information criterion (AICc) (Symonds & Moussalli, 2011; Whittingham, Stephens, Bradbury, & Freckleton, 2006). I selected the former to perform my preliminary analysis through the use of AICc and by selecting for the most relevant variables within each group. Although the model with the lowest AICc represents the best fit, model uncertainty can occur since models that differ by less than 2 AICc may not be statistically distinguishable (Anderson & Burnham 2002). If there are several models differing by less than 2 AICc from the best model, uncertainty occurs and a model averaging approach should be used (Symonds and Moussalli, 2011; Whittingham *et al*., 2006). This method takes into account all possible models, that differ by less than 2 AICs from the best one and averages them, thereby avoiding a possible parameter bias (reviewed by Whittingham *et al.,* 2006).

I conducted model averaging using the "dredge" function from the "MuMin" R package (Barton, 2014; see supplementary material for more details). For my analysis I compared all possible combination of models with mass and tarsus always included in the same model and never separated, as well as including time of sampling in all models concerning the oxidative stress markers. However, since the results from the most complete model with the lowest AICc value and the results from model averaging analysis gave similar conclusions for most questions, I chose, for simplicity, to present the estimates and p-values only from the model with more predictors within the range of 2 AICc. P values were computed using likelihood ratio tests (LRT) through the chi-squared distribution provided by the function "drop1" contained in the "stats" R package. All predictors were rescaled to a mean of 0 and a SD of 1 so that all estimates (and averaged estimates in the case of the model averaging analysis see supplement material) are comparable.

All analysis were conducted using R v. 3.2.5 (R Core Team 2016).

# Results

## Oxidative status and dominance

The ROMs concentration obtained was 0.656 ± 0.449 SD mg H2O2/dl (ranging from 0.103 – 2.641 mg H2O2/dl) and OXY concentration was 196.71 ± 55.023 SD HClO ml (ranging from 84.060 – 382.080 HClO ml). The ratio value calculated was 3.395 ± 1.970 SD (ranging from 0.392 – 10.797). Variation of ROMs concentration was significantly associated with the interaction between dominance and sex, with ROMs increasing with individual dominance score only in females, but not in males (β=-2.620 ± 0.936, df=1, LRT= 7.551, p= 0.006, Table 2, Fig. 3). This predictor was still significant after removing the three most extreme data points (see Fig. 3). There were no significant differences in ROMs levels between sexes (LMM after removing the interaction: β=-0.040 ± 0.234, df=1, LRT= 0.0287, p= 0.866). The interaction between dominance and age had a marginally significant effect, indicating that older birds with higher ROMs levels had lower dominance ranks (β=-0.930 ± 0.478, df=1, LRT= 3.616, p= 0.057, Table 2). When tested as single term, age had a significantly positive effect on ROMs (LMM after removing the interaction: β=0.214 ± 0.100, df=1, LRT= 4.430, p= 0.0353, Fig. 4). Body condition had no effect on ROMs, with mass being present in the most complex model (β=-0.103 ± 0.097, df=1, LRT= 1.109, p= 0.295, Table 2) and there was a positive effect of tarsus length on ROMs (β=0.248 ± 0.097, df=1, LRT= 6.350, p= 0.012, Table 2). The interaction between age and sex, although present in the most complete model, was not significant (β=0.533 ± 0.289, df=1, LRT= 3.221, p= 0.073, Table 2). The time elapsed between capture and sampling was positively related with ROMs concentration (β =0.160 ± 0.094, df=1, LRT=5.054, p= 0.025, Table 2).

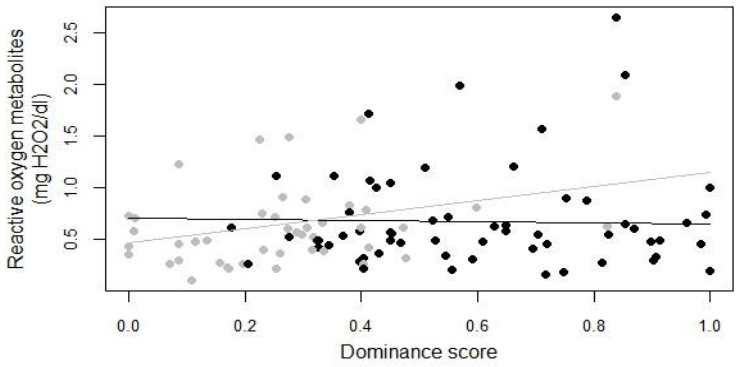


**Fig. 3** Relationship between the standardized David's score and the amount of reactive oxygen metabolites for each sex group for a total of 105 individuals (61 males and 44 females). Males are in black and females in grey. The lines represent predicted values estimated from the most complex model (Table 2).

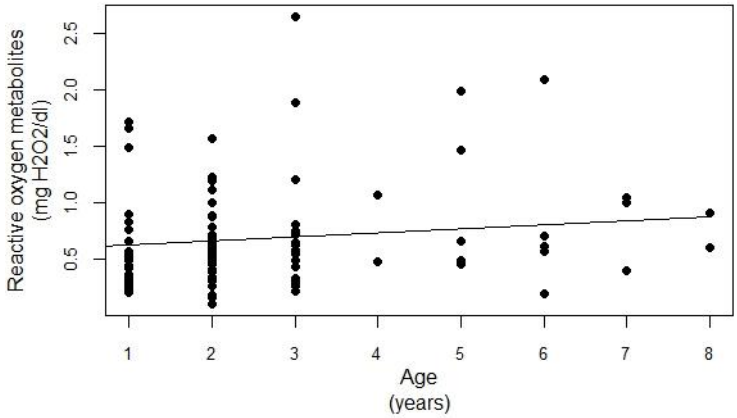


**Fig. 4:** Relationship between the amount of reactive oxygen metabolites (ROMs) and minimum age in years for a total of 105 individuals (61 males and 44 females). The line represents the predicted values estimated from the most complex model (Table 2).

Variation in individual OXY levels was positively related to sex, with males showing higher values of non-specific antioxidant defences (β= 0.364 ± 0.179, df=1, LRT= 4.049, p= 0.044, Table 2, Fig. 5). Although present in the most complex model, colony size did not significantly explained OXY variation (β= 0.208 ± 0.109, LRT= 3.244, df=1, p= 0.082, Table 2). As with ROMs, time of sampling was also positively related to OXY levels (β= 0.264 ± 0.135, LRT= 8.440, df=1, p= 0.004, Table 2). Age and dominance, body condition were either not present in the best models or when present were not significant (p>0.05, Table 2).

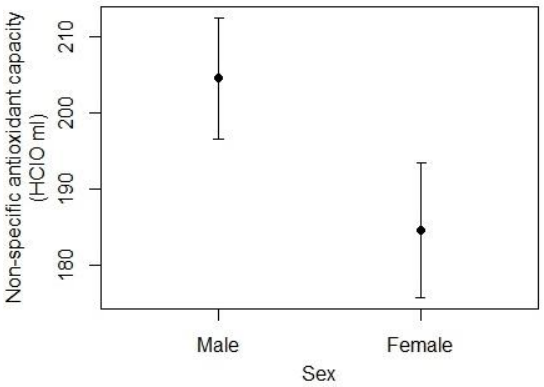


**Fig. 5:** Relationship between the non-specific antioxidant capacity (OXY) and sex (model estimates ± SE from the most complete model, Table 2) for a total of 105 individuals (61 males and 44 females).

The ratio between ROMs and OXY showed similar results to the ones found for ROMs variation, with the interactions between dominance and sex being significant, with more dominant females having a higher discrepancy between ROMs and OXY levels (β= -2.870 ± 0.967, df=1, LRT= 8.461, p= 0.004, Table 2, Fig. 6). The interaction between sex and age was marginally significant, with older males having higher ratio values than younger males (β= 0.574 ± 0.298, df=1, LRT= 3.623, p= 0.057, Table 2). Body condition did not explain individual oxidative stress, although mass was present in the most complex model (β= -0.039 ± 0.099, df=1, LRT= 0.157, p= 0.691, Table 2). Tarsus was the only term found to be significantly related to the ROM/OXY ratio (β= 0.227 ± 0.100, df=1, LRT= 5.053, p= 0.025, Table 2). All the remaining variables were not present in the best models or were not significant in the most complex one (p>0.05, Table 2).

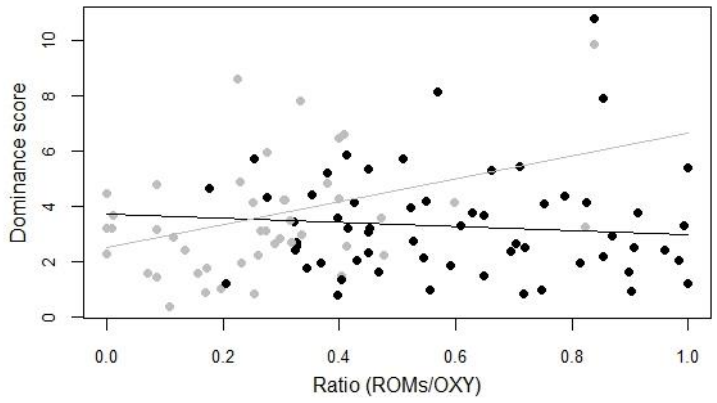


**Fig. 6** Relationship between the standardized David's score and the ratio (ROMs/OXY) for each sex group for a total of 105 individuals (61 males and 44 females). Males are in black and females in grey. The lines represent the predicted values estimated from the most complex model (Table 2).

## Oxidative status and bib size

Although included in the most complete model, bib size was not significantly related with ROMs (β=-0.112 ± 0.097, df=1, LRT= 1.323, p= 0.249, Table 3) or OXY (β=-0.033±0.100, df=1, LRT= 0.083, p= 0.742, Table 3). The quadratic bib size term was not present in the best models for both analyses. Regarding the ratio ROM/OXY, neither bib size nor the quadratic term were present in the best models.

**Table 2**. Results from the LMMs examining the effect of dominance in individual oxidative status components in the sociable weaver. Separated models for reactive oxygen metabolites (ROMs), non-specific antioxidant capacity (OXY) and ratio between markers for 105 individuals (61 males and 44 females) are represented. Estimates and p-values are presented for the variables present in the most complex model within the range of 2 AICc.

| | **ROMs** | | | **OXY** | | | **Ratio (ROM/OXY)** | | |
|---|---|---|---|---|---|---|---|---|---|
| | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* |
| *Final model* | | | | | | | | | |
| Intercept | -0.503 ± 0.240 | | | -0.222 ± 0.161 | | | -0.479 ± 0.244 | | |
| Dominance score (DSs)[1] | 2.235 ± 0.725 | - | - | x | x | x | 2.458 ± 0.738 | - | - |
| Age[2] | 0.501 ± 0.178 | - | - | x | x | x | 0.320 ± 0.181 | - | - |
| Sex[3] | 1.220 ± 0.454 | - | - | 0.364 ± 0.179 | 4.049 | **0.044** | 0.989 ± 0.462 | - | - |
| Mass | -0.103 ± 0.097 | 1.109 | 0.295 | x | x | x | -0.039 ± 0.099 | 0.157 | 0.691 |
| Tarsus | 0.248 ± 0.097 | 6.350 | **0.012** | x | x | x | 0.227 ± 0.100 | 5.053 | **0.025** |
| Colony size | x | x | x | 0.208 ± 0.109 | 3.244 | 0.082 | x | x | x |
| ST[4] | 0.216 ± 0.094 | 5.054 | **0.025** | 0.264 ± 0.135 | 8.440 | **0.004** | 0.147 ± 0.010 | 2.286 | 0.130 |
| Sex[3] * Age[2] | 0.533 ± 0.289 | 3.221 | 0.073 | x | x | x | 0.574 ± 0.298 | 3.623 | 0.057 |
| DSs[1] * Sex[3] | -2.620 ± 0.936 | 7.551 | **0.006** | x | x | x | -2.870 ± 0.967 | 8.461 | **0.004** |
| DSs[1] * Age[2] | -0.930 ± 0.478 | 3.616 | 0.057 | x | x | x | -0.796 ± 0.493 | 2.573 | 0.109 |
| DSs[1] * Sex[3] * Age[2] | x | x | x | x | x | x | x | x | x |

Reference categories: [1.] standardized David's score, [2.] minimum age in years, [3.] male, [4.] sampling time, [X.] variable not present in the most complex model within the range of 2 AICc.

**Table 3**. Results from the LMMs examining the effect of bib size in individual oxidative status components in the sociable weaver. Separated models for reactive oxygen metabolites (ROMs), non-specific antioxidant capacity (OXY) and ratio between markers for 87 individuals (52 males and 35 females) are represented. Estimates and p-values are presented for the variables present in the most complex model within the range of 2 AICc. Both linear and quadratic terms were tested for bib size.

| | **ROMs** | | | **OXY** | | | **Ratio (ROM/OXY)** | | |
|---|---|---|---|---|---|---|---|---|---|
| | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* |
| *Final model* | | | | | | | | | |
| Intercept | -0.053 ± 0.096 | | | -0.373 ± 0.194 | | | -0.038 ± 0.102 | | |
| Bib size | -0.112 ± 0.097 | 1.323 | 0.249 | -0.033 ± 0.100 | 0.101 | 0.742 | x | x | x |
| Age[1] | 0.146 ± 0.101 | 2.067 | 0.150 | x | x | x | x | x | x |
| Sex[2] | x | x | x | 0.473 ± 0.198 | 5.512 | **0.019** | x | x | x |
| Mass | x | x | x | x | x | x | 0.004 ± 0.113 | 0.001 | 0.973 |
| Tarsus | x | x | x | x | x | x | 0.163 ± 0.113 | 2.054 | 0.152 |
| ST[4] | 0.393 ± 0.101 | 13.837 | **<0.001** | 0.279 ± 0.098 | 7.725 | **0.006** | 0.283 ± 0.102 | 7.391 | **0.007** |
| Sex[1] * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size * Sex[2] | x | x | x | x | x | x | x | x | x |
| Bib size * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size * Sex[1] * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Sex[2] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Sex[1] * Age[1] | x | x | x | x | x | x | x | x | x |

Reference categories: [1.] Minimum age in years, [2.]male, [4.] sampling time, ^[2.]quadratic term, [X] variable not present in the most complex model within the range of 2 AICc.

## Correlates of bib size

There were no significant differences in bib size between sexes (sex was not present in the best models, Table 4). There was a positive effect of body mass on bib size, suggesting an association between a good body condition and larger bibs (β= 0.029 ± 0.109, df=1, LRT= 6.82, p= 0.009, Table 4). Age and colony size were present in the most complex model but were not significant (p>0.05, Table 4).

**Table 4**. Results from the LMM examining the correlates of individual bib size in the sociable weaver. The total of 87 individuals (52 males and 35 females) are represented. Estimates and p-values are presented for the variables present in the most complex model within the range of 2 AICc.

| | **Bib size** | | |
|---|---|---|---|
| | Estimate ± SE | LRT | *p* |
| *Final model* | | | |
| Intercept | -0.034 ± 0.151 | | |
| Age[1] | 0.165 ± 0.095 | 2.889 | 0.089 |
| Sex[2] | x | x | x |
| Mass | 0.294 ± 0.109 | 6.872 | **0.009** |
| Tarsus | -0.024 ± 0.108 | 0.049 | 0.823 |
| Colony size | -0.134 ± 0.132 | 0.971 | 0.324 |
| Sex[1] * Age[1] | x | x | x |

Reference categories: [1.] Minimum age in years, [2.]male, [X.] variable not present in the most complex model within the range of 2 AICc.

## Dominance and bib size

The interaction bib size*sex was present in the most complex model, although it was not significant (β=-0.046 ± 0.039, df=1, LRT= 1.361, p= 0.244, Table 5). However, when conducting the analyses separately for males and females, there was a significant effect of bib size for females (β =0.064 ± 0.031, LRT=3.946, p=0.047, Table 5, Fig. 7) whereas it was not significant for males for which only age was positively related with dominance (β =0.146 ± 0.024 LRT=29.03, p<0.001, Table 5).

**Table 5**. Results from the LMMs examining the relation between dominance and bib size in the sociable weaver. Separated models for all individuals, only males and only females of 93 individuals (55 males and 38 females) are represented. Estimates and p-values are presented for the variables present in most complex model within the range of 2 AICc.

| **Dominance score** | ***For all individuals*** | | | ***Only males*** | | | ***Only females*** | | |
|---|---|---|---|---|---|---|---|---|---|
| | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* | Estimate ± SE | LRT | *p* |
| *Final model* | | | | | | | | | |
| Intercept | 0.277 ± 0.029 | | | 0.586 ± 0.024 | | | 0.277 ± 0.029 | | |
| Bib size | 0.065 ± 0.032 | - | - | 0.019 ± 0.022 | 0.751 | 0.386 | 0.064 ± 0.031 | 3.946 | **0.047** |
| Age[1] | -0.006 ± 0.028 | - | - | 0.146 ± 0.024 | 29.039 | **<0.001** | x | x | x |
| Sex[2] | 0.310 ± 0.028 | - | - | - | - | - | - | - | - |
| Mass | x | x | x | x | x | x | x | x | x |
| Tarsus | x | x | x | x | x | x | x | x | x |
| Colony size | x | x | x | x | x | x | x | x | x |
| Sex[1] * Age[1] | 0.152 ± 0.028 | 15.141 | **<0.001** | x | x | x | x | x | x |
| Bib size * Sex[2] | -0.046 ± 0.039 | 1.361 | 0.244 | - | - | - | - | - | - |
| Bib size * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size * Sex[1] * Age[1] | x | x | x | - | - | - | - | - | - |
| Bib size^[2] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Sex[2] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Age[1] | x | x | x | x | x | x | x | x | x |
| Bib size^[2] * Sex[1] * Age[1] | x | x | x | x | x | x | x | x | x |

Reference categories: [1.] Minimum age in years, [2.]male, ^[2.]quadratic term , [X.]variable not present in the most complex model within the range of 2 AICc.

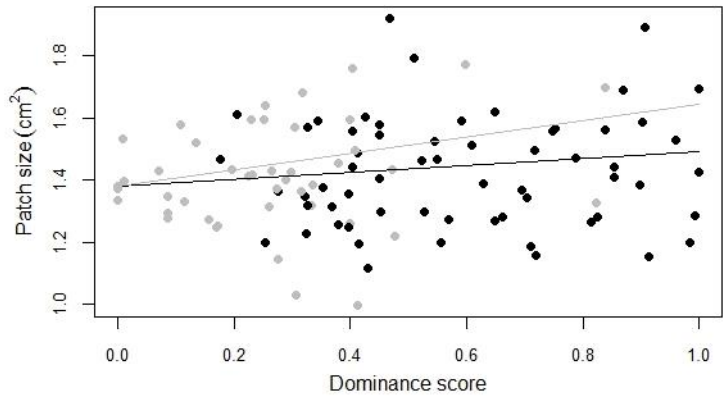


**Fig. 7** Relationship between individual's standardized David's score and bib size estimates for each sex group for a total of 93 individuals (55 males and 38 females). Males are in black and females in grey. The lines represent the predicted values estimated from the most complex model (Table 5).

Dominance was found significantly explained by the interaction between sex and age, with males dominant over females and with dominance rank increasing with age only in males (β=0.152 ± 0.028, df=1, LRT= 15.141, p= <0.001, Table 5).

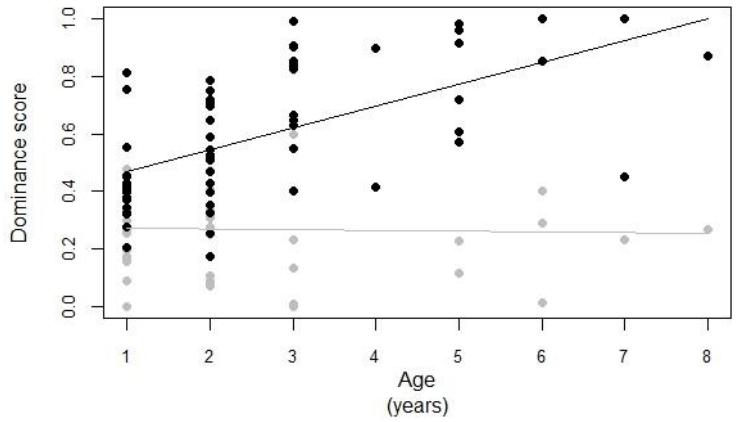


**Fig. 8** Relationship between the standardized David's score and age for each sex group for a total of 105 individuals (61 males and 44 females). Males are in black and females in grey. The lines represent the predicted values estimated from the most complex model (Table 5).

## Correlation between the two markers

ROMs levels were positively correlated with OXY levels (r=0.22, N=105, P=0.024). Although, when testing for separate sexes, this correlation in both markers was found only significant for males (males: r=0.29, N=61, P=0.022, females: r=0.07, N=44, P=0.670).

# Discussion

This study investigated possible physiological costs underlying social dominance. I found that oxidative stress, the physiological measure used here, may underpin a possible trade-off between the benefits of dominance and the associated physiological costs; however, this effect appears to occur only in females. The results obtained in this study also provided some indication of oxidative status being affected by the age of the individuals. There was no relationship between the two oxidative markers and bib size, even though bib size was related with dominance in females. Finally, males were dominant over females and dominance was found to be explained by age only in males.

The main results from this work suggest that, in the sociable weaver, rank-related differences in oxidative status only emerged in a sex-specific manner, with oxidative stress and damage increasing with dominance only in females. This increase of oxidative stress with dominance in females may be explained by the increase in aggressive and policing behaviour (through displacements and threatening) with rank, characteristics of both dominance acquisition and maintenance. Although females are subordinate to males (only 28% of females attain a medium or higher rank, compared to the almost 98% of males) and were less often observed engaging in aggressive interactions (only 13% of females initiated an aggressive interaction compared to the 87% initiated by males), this scarce fights to attain a higher rank within females may represent a higher cost and may be causing an over-production of ROS, ultimately translated in ROMs. This result suggests that females may be struggling to maintain redox homeostasis when attaining and maintaining a higher and demanding dominant position. From a physiological point of view, this may be explained by the sex-specific differences found in total antioxidant capacity with higher OXY levels found in males. A relation that was supported by the positive correlation between the markers in a sex-specific manner, where higher ROMs concentrations seem to be compensated by an increase in OXY defences only in males. This OXY compensatory effect may be failing to mitigate the dominance related ROS production in females.

The physiological costs associated with rank in females may be explained by hormonal changes, translated in the production of stress hormones (such as glucocorticoids) and an increase in circulating androgens (such as testosterone) with rank, both linked with aggressiveness (Wingfield *et al.* 1987; Creel *et al.* 2013) and signal maintenance (Lattin & Romero 2013; Bókony *et al.* 2008). These hormonal changes, which were previously

found to be positively correlated with rank (Creel 2005; Poisbleau *et al.* 2005), were also found to generate higher levels of ROS in other species (Alonso-Alvarez *et al.* 2007; Costantini *et al.* 2011). Although a similar scenario should also be expected for dominant males, the lack of rank-related differences in males in the levels of oxidant species may possibly be explained by the different reproductive strategies and/or the differential access to food resources experienced by the two sexes.

In fact, many species showed a similar relation between antioxidant capacity and sex (Isaksson 2013; Wegmann *et al*. 2015; Alonso-Alvarez *et al.* 2004; Beaulieu *et al.* 2014). One possible explanation for these sex differences in redox physiology found in this investigation may lie on the already reported sex-difference in physiology and reproductive strategies (Monaghan *et al.* 2009; Costantini 2008). These differences may affect the functioning of the antioxidant system and are commonly translated in a decrease in females' antioxidant defences, as a differential cost of reproduction itself (Monaghan *et al.* 2009; Costantini 2008). Cram *et al.* 2014, found that in the white-browed sparrow weaver *Plocepasser mahali* rank-related differences only emerged after the breeding season, a pattern linked with disparities in reproductive effort in a sex-specific manner. Resources allocated to reproduction, differential workload and energetic demands, followed by a differential pattern of circulating hormones (Haliwell & Gutteridge, 2007), can be linked with breeding effort and egg production, laying and incubation, and may therefore explain higher oxidation risk of females (Alonso-Alvarez *et al.*, 2004; Blount *et al.* 2000; Cram *et al.,* 2014; Travers, *et al.* 2010).). In the case of this study at the time of the sampling, 16 juveniles (with less than 3 months old) were captured suggesting that until recently breeding was still occurring in the study population and thus females might have been still recovering from the breeding effort.

Another possible and complementary explanation to the lower capacity of females to up-regulate the burden of dominance may be due to the expected differential access between sexes to resources, since males are dominant over females and are expected to exert some control in the access to resources such as food, which provides source of antioxidant-rich dietary resources (Catoni *et al.* 2008). This control over food may have been accentuated by the stronger 2015 El Niño dry season. Indeed, both markers are likely to be influenced by food quantity/quality, since several dietary oxidants and antioxidants can be detected by the assays *(Costantini 2010; Van de Crommenacker et al.* 2011). Therefore, this restricted access may be preventing females from pre-emptively regulate antioxidant protection depleted after reproduction, and return to the normal basal levels outside the breeding season, explaining the maintenance of the sex differences in antioxidant defences after the breeding season and at the time of

sampling. Although there's no significant differences in body condition between sexes, females may not have the same access to high-quality food as males, and simultaneously may need more energy and time to access this resources, a struggle that may have consequences in the oxidative status (Monaghan *et al.* 2009; Speakman *et al.* 2015) and explain the obtained results.

ROMs were also found to be positively related to age. Older sociable weavers seem to have an increased susceptibility to the environmental, social and/or physiological stressors than younger ones. This relationship with age has been found in several species (Martin & Grotewiel 2006) and has been explained either by a higher rate of ROS production associated with mitochondria dysfunctions, or with a higher intrinsic susceptibility of molecules to ROS as the result of an age related increase in the degree of unsaturation of the membrane polyunsaturated fatty acid and/or a decrease in oxidized molecules repair or excision mechanism (reviewed in Monaghan *et al.* 2009). A deleterious effect that may have an important role in the individual's fitness and other life-histories components, such as growth, reproduction, pathology and diseases' defence; this *per se* may ultimately relate to ageing (Martin & Grotewiel 2006). These findings that only ROMs levels, and not the antioxidant defences, were found to be related with age stands in accord with the cumulative evidence that lifespan and ageing may be affected by ROS production and susceptibility and not by differences in the antioxidant defences (Monaghan *et al*., 2009).

The higher levels of OXY found in males, and which were not related to mass and/or tarsus, can reflect an adaptive and compensatory response to physiological stress promoted by other stressors, rather than optimal health condition (Horak & Cohen 2010). Similarly, the lack of differences in antioxidant defences with rank, as well with body condition, counters the hypothesis that dominants are in better condition because of intrinsic quality or better competitive ability. A comparable result was also found in the white-browed sparrow weaver (Cram *et al*., 2014) and in the mandrill (Beaulieu et al. 2014), with no differences in the antioxidant capacity between dominant breeders and subordinate non-breeders.

Finally, the lack of differences found for both sexes in the OXY defences with rank may also be due to the lower sensitivity of the OXY assay in detecting environmental changes when compared with the ROMs assay, due to the endogenously synthetized antioxidants (Monaghan *et al*., 2009; Van de Crommenacker *et al*., 2011).

Inferring variation in the complex oxidative status through only looking at oxidant or antioxidant response may be misleading (Monaghan *et al.* 2009; Costantini & Verhulst

2009). The ratio between ROMs and OXY allowed to assess both sides of the oxidative balance and to infer if there was indeed oxidative damage mediating the costs of dominance in the sociable weaver. The higher the value of this ratio, the higher the differences between the oxidant and antioxidant components (with oxidants higher than antioxidant defences), thus suggesting an imbalance that may translate in oxidative stress, and ultimately, in oxidative damage. The fact that this ratio was also found to be explained by dominance in a sex-specific manner, with dominant females showing higher discrepancies between markers, stands in accord with the hypothesis that females are struggling to upregulate the increase production of ROS associated with attaining and maintaining an higher dominant and demanding position promoted by the depleted antioxidant defences in comparison with males.

Nevertheless, to demonstrate my assumptions regarding oxidative status and dominance, future challenges may involve assessing the individual hormonal profile associated with aggressiveness or dominance, which may explain the rank-related differences found in oxidants production on females. Additionally, both the individual breeding effort and the possible differential access to food resources should be accessed to demonstrate a potential link to the sex-differences found in antioxidant defences.

Contrary to my expectations, bib size was only related to individual body condition and not with individual oxidative status components. This relation between body condition and melanin-based plumage colouration was previously found in other species (Nakagawa *et al.* 2007), and could be linked with signalling honesty (e.g. only individuals in good condition are able to entail the bib production and display costs, Zahavi *et al.* 1999). Roulin (2015) suggested that a colour trait could signal quality not only because it is costly to produce, but because of pleiotropy and the genetic link between colouration and body condition. Therefore, the production of melanin pigments may require genes that affect body condition and, ultimately, physiology and oxidative status (Roulin, 2015). For instance, in the great tit *Parus major*, the melanin based black stripe was in fact signalling body condition through the individual capacity to manage oxidative stress, since the production of the colour trait was at the expense of the glutathione (a key intracellular antioxidant) and the resistance to oxidative stress (Galván & Alonso-Alvarez 2008). However, this genetic link between the expression of pleiotropic melanogenic genes may vary depending on the environmental conditions or life stages (Roulin 2015). Therefore, one possible explanation for the lack of impact in individual oxidative status detected is that the expression of the sociable weaver's bib may be context-dependent and can be influenced by the environment and by body condition only at the specific times when these structures were produced (Roulin 2015). If melanin pigments are costly

by requiring specific nutrients or energy only at time of production, oxidative differences may be only significant in this species in juveniles (when the bib starts to develop) or during moulting. Although this remains speculative and future work on the physiological costs associated with bib production in this species should be focused on juveniles.

Secondly, and relative to the lack of social costs associated with bib display and linked impacts on oxidative status, expression and assessment of the bib may be cheaper in association with the social context. A scenario that may be supported by the fact that status signalling lost its relevance for dominance signalling in males. The dry season that occurred this year seems to have shaped the frequency and type of interactions. Medium size bibs were not associated with increased aggression and assessment as expected (Rat *et al.* 2015; Maynard Smith *et al.* 1988), and there was an increase in the engagement in aggressive fights between males (increased almost 13%, when compared with Rat *et al.* 2015), while avoidance behaviour and the active exclusion towards conspecifics decreased at the plate (see Fig.9 for an illustrative comparison). This may be promoted by a hunger effect in the willingness to win contests and access the feeder, which may alter the contest outcomes, dominance relations and signalling function (Andersson & Åhlund 1991). In fact, the inter-annual variation in the relationship between survival and bib size suggests that the environment, and more specifically rainfall should affect bib size variation, due to its effects in food availability and the in level of competition for food (Acker *et al.* 2015). As a result, this environmental fluctuation may affect signalling ability and assessment (Acker *et al.* 2015). This type of scenario is also described in the literature as the “desperados” effect (Grafen 1987), where individuals escalate in highly costly contests for the scarce resources when the costs of not eating surpluses the costs of the agonistic interaction. This may possibly explain why expected subordinates with small bibs may be winning expected dominants with larger bibs, distorting the signalling function of the bib in males. Decreased food availability may also explain why females showed a slight increase in trying to access the feeder (6% more females presence recorded compared with Rat *et al.,* 2015). However there was an effect of bib size on dominance of females. In contrast with males, there was no increased aggression in females compared to Rat *et al.,* (2015) and therefore maintenance of the signalling function may still be used to avoid escalated fights in females. To test these possible explanations, future challenges should look at long-term data to test if environmental conditions could have an effect on how dominance status is related with badge size and, thus, with signal assessment and possible costs.

Although not feasible due to the difficulties of sampling at the field (Salin *et al.* 2015) and the limited volume of plasma available, multiple markers for both oxidative damage and

antioxidant defences are needed to get better picture of the relation between oxidative status and dominance in the sociable weaver. The amount of oxidized molecules measured in a tissue may depend on intrinsic susceptibility of the molecule to oxidization by ROS (as stated before, the degree of peroxidization of fatty acids) and the efficiency of the repair, degradation and turnover of the oxidized molecules by the antioxidant machinery, processes that could not be assumed by focusing only on plasma antioxidant capacity or ROS quantity (Pamplona & Costantini 2011; Rey *et al.* 2016). Previous studies on mammals demonstrate low or no correlation between different oxidative markers, enlightening the necessity for the assessment of multiple markers and to assess multiple antioxidant mechanisms (Christensen *et al.* 2015). Furthermore, care must be taken when generalizing from these results, as each marker may reflect different and potentially uncoupled biochemical processes in different tissues and different species (Speakman *et al.* 2015; Christensen *et al.* 2015; Monaghan *et al.* 2009).

Finally, the time of sampling, as a proxy between capture and the completion of blood sampling, although lessened as possible, and contrary to previous findings suggesting that acute increases in stress hormones linked with handling or containment do not induce immediate, short-term changes in ROMs levels and/or OXY (Costantini *et al.* 2007; Costantini *et al.* 2008; Herborn *et al.* 2011) was highly significant in determining the variation of both the study markers. Indeed, handling and containment confront individuals with a multitude of stressors that may challenge internal homeostasis and, consequently, oxidative stress. For example, acute increases in corticosterone, a glucocorticoid proved to be linked with stress response, was found to be linked with the increase of both the production of ROS and the enhance the antioxidant capacity in a compensatory manner, in broiler chickens *Gallus gallus domesticus* (Lin *et al.* 2004), a pattern similar to the one observed in my results. Although in normal free-ranging conditions animals have time to recover, when sampling we may only measure the immediate changes (e.g. the acute oxidative state) and not the true physiological state of the individual (e.g. the chronic oxidative state) (Chung *et al.* 2013; Seet *et al.* 2011). One possibility to mitigate this deleterious effects on the individuals and to have more clear responses would be to employ a non-invasive approach in the measurement of multi-tissue oxidative stress, a need for innovation already reported to be accomplished in the future through the considerable information that can be gathered in feathers, hair, urine and faecal samples (Speakman *et al.* 2015).

To my knowledge, this was the first study to examine the link between oxidative status and an individual's position in the social hierarchy (i.e. dominance rank), however other studies have looked to this relation of oxidative status and dominance through breeding

status (Beaulieu *et al.,* 2014; Cram *et al.*, 2014). Together the findings of this work suggest that indeed dominance is underpinned by oxidative stress in a sex-related manner, with dominant females showing increased levels of oxidative damage than their subordinates. Dominant females seem to not be able to pre-empt the oxidative costs of the social competition, a limitation that can be explained by the sex-specific variation in oxidant defences. Males show higher antioxidant levels than females, independent of the rank, a pattern than can be explained either by the differential access to the scarce food resources or by the different reproductive strategies and effort. Therefore these findings do reflect sex differences in behaviour, possibly related with food intake and/or reproductive physiology that are selecting different adaptive strategies that may be influencing oxidative status through individual life-histories. On the contrary, bib size was not related with any individual redox component, although showing an important signalling function for females and an important relation with body condition, supporting the honesty of this signal. Furthermore, in addition to Cram *et al.* (2014) results and the endocrine stress rank-related differences reported by Creel (2001), the present study adds to the accumulating evidence that social dominance in cooperative species may entail trade-offs between advantages and hidden physiological costs.


**Fig. 9** Sociable weavers interacting at feeder in consecutive years: A) 2013, B) 2014 and C) 2015.

# References

**Acker, P., Grégoire, A., Rat, M., Spottiswoode, C. N., van Dijk, R. E., Paquet, M., … Doutrelant, C. (2015).** Disruptive viability selection on a black plumage trait associated with dominance. *Journal of Evolutionary Biology*, *28*(11), 2027–2041.

**Alberti, A., Bolognini, L., Macciantelli, D., & Caratelli, M. (2000).** The radical cation of N,N-diethyl-para-phenylendiamine: A possible indicator of oxidative stress in biological samples. *Research on Chemical Intermediates*, *26*(3), 253–267.

**Alonso-Alvarez, C., Bertrand, S., Devevey, G., Prost, J., Faivre, B., & Sorci, G. (2004).** Increased susceptibility to oxidative stress as a proximate cost of reproduction. *Ecology Letters,* 7(5), 363-368.

**Alonso-Alvarez, C., Bertrand, S., Faivre, B., Chastel, O., & Sorci, G. (2007).** Testosterone and oxidative stress: the oxidation handicap hypothesis. *Proceedings. Biological Sciences / The Royal Society*, *274*(1611), 819–25.

**Altizer, S., Nunn, C. L., Thrall, P. H., Gittleman, J. L., Antonovics, J., Cunningham, A. A., … Pulliam, J. R. C. (2003).** Social Organization and Parasite Risk in Mammals: Integrating Theory and Empirical Studies. *Annual Review of Ecology, Evolution, and Systematics*, *34*, 517–547.

**Anderson, D. R., & Burnham, K. P. (2002).** Avoiding Pitfalls When Using Information-Theoretic Methods. *The Journal of Wildlife Management*, *66*(3), 912.

**Andersson, S., & Åhlund, M. (1991).** Hunger affects dominance among strangers in house sparrows. *Animal Behaviour,* 41(5), 895–897

**Ang, T. Z., & Manica, A. (2010).** Aggression, segregation and stability in a dominance hierarchy. *Proceedings. Biological Sciences / The Royal Society*, *277*(1686), 1337–1343.

**Arnott, S. A., Chiba, S., & Conover, D. O. (2006).** Evolution of intrinsic growth rate: metabolic costs drive trade-offs between growth and swimming performance in Menidia menidia. *Evolution; International Journal of Organic Evolution*, *60*(6), 1269–1278.

**Babior, B. M., Kipnes, R. S., & Curnutte, J. T. (1973).** Biological defense mechanisms. The production by leukocytes of superoxide, a potential bactericidal agent. *The Journal of Clinical Investigation*, *52*(3), 741–4.

**Barton, K. (2014).** MuMIn: Multi-model inference. http://cran.r-project.org/package=MuMIn.

**Bates, D., Maechler, M., Bolker, B. M., & Walker, S. (2014).** lme4: Linear mixed- effects models using Eigen and S4. http://cran.r-project.org/package=lme4.

**Beaulieu, M., Mboumba, S., Willaume, E., Kappeler, P. M., & Charpentier, M. J. E. (2014).** The oxidative cost of unstable social dominance. *The Journal of Experimental Biology*, *217*(15), 2629–2632.

**Birkhead, T. R., Møller, A. P. (1992).** *Sperm competition in birds.* Academic Press.

**Blount, Houston, & Møller. (2000).** Why egg yolk is yellow. *Trends in Ecology & Evolution*, *15*(2), 47–49.

**Bókony, V., Garamszegi, L. Z., Hirschenhauser, K., & Liker, A. (2008).** Testosterone and melanin-based black plumage coloration: a comparative study. *Behavioral Ecology and Sociobiology*, *62*(8), 1229–1238.

**Catoni, C., Peters, A., & Schaefer, H. M. (2008).** Life history trade-offs are influenced by the diversity, availability and interactions of dietary antioxidants. *Animal Behaviour*, *76*(4), 1107–1119.

**Chancellor, R. L., & Isbell, L. A. (2009).** Food site residence time and female competitive relationships in wild gray-cheeked mangabeys (Lophocebus albigena). *Behavioral Ecology and Sociobiology*, *63*(10), 1447–1458.

**Chiarati, E., Canestrari, D., Vila, M., Vera, R., & Baglione, V. (2011).** Nepotistic access to food resources in cooperatively breeding carrion crows. *Behavioral Ecology and Sociobiology*, *65*(9), 1791–1800.

**Christensen, L. L., Selman, C., Blount, J. D., Pilkington, J. G., Watt, K. A., Pemberton, J. M., … Nussey, D. H. (2015).** Plasma markers of oxidative stress are uncorrelated in a wild mammal. *Ecology and Evolution*, *5*(21), 5096–5108.

**Chung, M. L. S., Lee, K. Y. E., & Lee, C. Y. J. (2013).** Profiling of oxidized lipid products of marine fish under acute oxidative stress. *Food and Chemical Toxicology*, *53*, 205–213.

**Clutton-Brock, T. H., Hodge, S. J., Spong, G., Russell, A. F., Jordan, N. R., Bennett, N. C., … Manser, M. B. (2006).** Intrasexual competition and sexual selection in cooperative mammals. *Nature*, *444*(7122), 1065–1068.

**Constantini, N., & Hackney, A. C. (2013).** *Endocrinology of Physical Activity and Sport: Second Edition*. New York: Humana Press.

**Costantini, D. (2008).** Oxidative stress in ecology and evolution: Lessons from avian studies. *Ecology Letters,* 11(11), 1238-1251.

**Costantini, D. (2010).** Complex trade-offs in the pigeon (Columba livia): egg antioxidant capacity and female serum oxidative status in relation to diet quality. *Journal of Comparative Physiology B*, *180*(5), 731–739.

**Costantini, D., Cardinale, M., & Carere, C. (2007).** Oxidative damage and anti-oxidant capacity in two migratory bird species at a stop-over site. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, *144*(4), 363–371.

**Costantini, D., Casagrande, S., De Filippis, S., Brambilla, G., Fanfani, A., Tagliavini, J., & Dell'Omo, G. (2006b).** Correlates of oxidative stress in wild kestrel nestlings (Falco tinnunculus). *Journal of Comparative Physiology B*, *176*(4), 329–337.

**Costantini, D., Dell'ariccia, G., & Lipp, H.-P. (2008).** Long flights and age affect oxidative status of homing pigeons (Columba livia). *The Journal of Experimental Biology*, *211*(Pt 3), 377–381.

**Costantini, D., & Dell'Omo, G. (2006).** Effects of T-cell-mediated immune response on avian oxidative stress. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, *145*(1), 137–142.

**Costantini, D., & Dell'Omo, G. (2015).** Oxidative stress predicts long-term resight probability and reproductive success in Scopoli's shearwater (*Calonectris diomedea*). *Conservation Physiology*, *3*(1), cov024.

**Costantini, D., Marasco, V., & Møller, A. P. (2011).** A meta-analysis of glucocorticoids as modulators of oxidative stress in vertebrates. *Journal of Comparative Physiology B*, 181(4), 447-456

**Costantini, D., Metcalfe, N. B., & Monaghan, P. (2010).** Ecological processes in a hormetic framework, *13*(11), 1435–47.

**Costantini, D., & Verhulst, S. (2009).** Does high antioxidant capacity indicate low oxidative stress? *Functional Ecology*, *23*(3), 506–509.

**Costantini, D. (2016).** Oxidative stress ecology and the d-ROMs test: facts, misfacts and an appraisal of a decade's work. *Behavioral Ecology and Sociobiology,* 70(5), 809-820.

**Côté, S. D., & Festa-Bianchet, M. (2001).** Reproductive success in female mountain goats: the influence of age and social rank. *Animal Behaviour*, *62*(1), 173–181.

**Covas, R., Dalecky, A., Caizergues, A., & Doutrelant, C. (2006).** Kin associations and direct vs indirect fitness benefits in colonial cooperatively breeding sociable weavers Philetairus socius. *Behavioral Ecology and Sociobiology*, *60*(3), 323–331.

**Covas, R., du Plessis, M. a., & Doutrelant, C. (2008).** Helpers in colonial cooperatively breeding sociable weavers Philetairus socius contribute to buffer the effects of adverse breeding conditions. *Behavioral Ecology and Sociobiology*, *63*(1), 103–112.

**Cram, D. L., Blount, J. D., & Young, A. J. (2014).** Oxidative status and social dominance in a wild cooperative breeder. *Functional Ecology*, 29(2), 229-238.

**Cram, D. L., Blount, J. D., & Young, A. J. (2015).** The oxidative costs of reproduction are group-size dependent in a wild cooperative breeder. *Proceedings of the Royal Society of London B: Biological Sciences*, *282*(1819), 2015-2031.

**Creel, S. (2001).** Social dominance and stress hormones. *Trends in Ecology & Evolution*. 16(9), 491-497.

**Creel, S. (2005).** Dominance, Aggression, and Glucocorticoid Levels in Social Carnivores. *Journal of Mammalogy*, *86*(2), 255-264.

**Creel, S., Dantzer, B., Goymann, W., & Rubenstein, D. R. (2013).** The ecology of stress: Effects of the social environment. *Functional Ecology*, *27*(1), 66–80.

**David, H. A. (1987).** Ranking from unbalanced paired-comparison data. *Biometrika*, *74*(2), 432–436.

**De Vries, H., Stevens, J. M. G., & Vervaecke, H. (2006).** Measuring and testing the steepness of dominance hierarchies. *Animal Behaviour*, *71*(3), 585–592.

**Ellison, P. T. (2003).** Energetics and reproductive effort. *American Journal of Human Biology*, *15*(3), 342–351.

**Finkel, T., & Holbrook, N. J. (2000).** Oxidants, oxidative stress and the biology of ageing. *Nature*, *408*(6809), 239–247.

**Galván, I., & Alonso-Alvarez, C. (2008).** An Intracellular Antioxidant Determines the Expression of a Melanin-Based Signal in a Bird. *PLoS ONE*, *3*(10), e3335.

**Gammell, M. P., & de Vries, H. (2003).** David's score: a more appropriate dominance ranking method than Clutton-Brock et al.'s index. *Animal Behaviour*, *66*(3), 601–605.

**Geiger, S., Le Vaillant, M., Lebard, T., Reichert, S., Stier, A., Le Maho, Y., & Criscuolo, F. (2012).** Catching-up but telomere loss: Half-opening the black box of growth and ageing trade-off in wild king penguin chicks. *Molecular Ecology*, *21*(6), 1500–1510.

**Georgiev, A. V., Muehlenbein, M. P., Prall, S. P., Emery Thompson, M., & Maestripieri, D. (2015).** Male quality, dominance rank, and mating success in free-ranging rhesus macaques. *Behavioral Ecology*, *26*(3), 763–772.

**Gerald, M. S. (2001).** Primate colour predicts social status and aggressive outcome. *Animal Behaviour*, *61*(3), 559–566.

**Grafen, A. (1987).** The logic of divisively asymmetric contests: respect for ownership and the desperado effect. *Animal Behaviour*, *35*(2), 462–467.

**Griffiths, R., Double, M. C., Orr, K., & Dawson, R. J. (1998).** A DNA test to sex most birds. *Molecular Ecology*, *7*(8), 1071–1075.

**Guthery, F. S., Brennan, L. A., Peterson, M. J., & Lusk, J. J. (2005).** Information theory in wildlife science: critique and viewpoint. *Journal of Wildlife Management*, *69*(2), 457–465.

**Halliwell, B., Gutteridge, J.-C. (2007).** *Free Radicals in Biology and Medicine.* Oxford: Oxford University Press.

**Harrell, F. E., & Dupont, C. (2016).** Hmisc: Harrell miscellaneous. http://cran.rproject.org/package= Hmisc.

**Herborn, K. A., Coffey, J., Larcombe, S. D., Alexander, L., & Arnold, K. E. (2011).** Oxidative profile varies with personality in European greenfinches. *The Journal of Experimental Biology*, *214*(Pt 10), 1732–1739.

**Horak, P., & Cohen, A. (2010).** How to measure oxidative stress in an ecological context: Methodological and statistical issues. *Functional Ecology*, *24*(5), 960–970.

**Hughes, N. F. (1992).** Ranking of Feeding Positions by Drift-Feeding Arctic Grayling (Thymallus arcticus) in Dominance Hierarchies. *Canadian Journal of Fisheries and Aquatic Sciences*, *49*(10), 1994–1998.

**Isaksson, C. (2013).** Opposing effects on glutathione and reactive oxygen metabolites of sex, habitat, and spring date, but no effect of increased breeding density in great tits (Parus major). *Ecology and Evolution*, *3*(8), 2730–2738.

**Kappeler, P. M., & van Schaik, C. P. (2002).** Evolution of Primate Social Systems. *International Journal of Primatology*, *23*(4), 707–740.

**Kilk, K., Meitern, R., Härmson, O., Soomets, U., & Hõrak, P. (2014).** Assessment of oxidative stress in serum by d-ROMs test. *Free Radical Research*, *48*(8), 883–889.

**Koenig, W. D., Walters, E. L., & Haydock, J. (2011).** Fitness consequences of within-brood dominance in the cooperatively breeding acorn woodpecker. *Behavioral Ecology and Sociobiology*, *65*(12), 2229–2238.

**Lardy, S., Allainé, D., Bonenfant, C., & Cohas, A. (2015).** Sex-specific determinants of fitness in a social mammal. *Ecology*, *96*(11), 2947–2959.

**Lardy, S., Rey, B., Salin, K., Voituron, Y., & Cohas, A. (2016).** Beneficial effects of group size on oxidative balance in a wild cooperative breeder. *Behavioral Ecology*, arw114.

**Lattin, C. R., & Romero, L. M. (2013).** The size of a melanin-based plumage ornament correlates with glucocorticoid receptor concentrations in the skin of that ornament. *Biology Letters*, *9*(5), 20130440.

**Laubach, Z. M., Blumstein, D. T., Romero, L. M., Sampson, G., & Foufopoulos, J. (2013).** Are white-crowned sparrow badges reliable signals? *Behavioral Ecology and Sociobiology*, *67*(3), 481–492.

**Lin, H., Decuypere, E., & Buyse, J. (2004).** Oxidative stress induced by corticosterone administration in broiler chickens (Gallus gallus domesticus): 1.Chronic exposure. *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*, 139(4), 737–744.

**Lindschinger, M., Nadlinger, K., Adelwöhrer, N., Holweg, K., Wögerbauer, M., Birkmayer, J., … Wonisch, W. (2004).** Oxidative stress: potential of distinct peroxide determination systems. *Clinical Chemistry and Laboratory Medicine*, *42*(8), 907-914.

**Maclean., G, L. (1973).** The sociable weaver, part 3: breeding biology and moult. *Ostrich*, 44(3-4), 219–240.

**Martin, I., & Grotewiel, M. S. (2006).** Oxidative damage and age-related functional declines. *Mechanisms of Ageing and Development*, *127*(5), 411–423.

**Maynard Smith, J., Harper, D. G., & Brookfield, J. F. Y. (1988).** The evolution of aggression: can selection generate variability? *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences*, *319*(1196), 557–570.

**Maynard-Smith, J. & Harper, D. 2003.** *Animal Signals.* Oxford: Oxford University Press

**Monaghan, P., Metcalfe, N. B., & Torres, R. (2009).** Oxidative stress as a mediator of life history trade-offs: Mechanisms, measurements and interpretation. *Ecology Letters*, *12*(1), 75–92.

**Nakagawa, S., Ockendon, N., Gillespie, D. O. S., Hatchwell, B. J., & Burke, T. (2007).** Assessing the function of house sparrows' bib size using a flexible meta-analysis method. *Behavioral Ecology*, *18*(5), 831–840.

**Nelson-Flower, M. J., Hockey, P. A. R., O'Ryan, C., Raihani, N. J., du Plessis, M. A., & Ridley, A. R. (2011).** Monogamous dominant pairs monopolize reproduction in the cooperatively breeding pied babbler. *Behavioral Ecology*, *22*(3), 559–565.

**Nelson-flower, M. J., & Ridley, A. R. (2015).** Male-male competition is not costly to dominant males in a cooperatively breeding bird. *Behavioral Ecology and Sociobiology*, *69*(12), 1997–2004.

**Nemoto, S., Takeda, K., Yu, Z. X., Ferrans, V. J., & Finkel, T. (2000).** Role for mitochondrial oxidants as regulators of cellular metabolism. *Molecular and Cellular Biology*, *20*(19), 7311–7318.

**Pamplona, R., & Costantini, D. (2011).** Molecular and structural antioxidant defenses against oxidative stress in animals. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology*, *301*(4), R843-R863.

**Poiani, A., Goldsmith, A. R., & Evans, M. R. (2000).** Ectoparasites of house sparrows (Passer domesticus): an experimental test of the immunocompetence handicap hypothesis and a new model. *Behavioral Ecology and Sociobiology*, *47*(4), 230–242.

**Poisbleau, M., Fritz, H., Guillemain, M., & Lacroix, A. (2005).** Testosterone and Linear Social Dominance Status in Captive Male Dabbling Ducks in Winter. *Ethology*, *111*(5), 493–509.

**Poisbleau, M., Fritz, H., Valeix, M., Perroi, P.-Y., Dalloyau, S., & Lambrechts, M. M. (2006).** Social dominance correlates and family status in wintering dark-bellied brent geese, Branta bernicla bernicla. *Animal Behaviour*, *71*(6), 1351–1358.

**Rammal, H., Bouayed, J., & Soulimani, R. (2010).** A direct relationship between aggressive behavior in the resident/intruder test and cell oxidative status in adult male mice. *European Journal of Pharmacology*, *627*(1-3), 173–176.

**Rat, M., van Dijk, R. E., Paquet, M., Silva, L., Doutrelant, C., & .Covas, R. (submitted)**. Multiple benefits of dominance status in a highly social bird. *Behavioral Ecology.*

**Rat, M. (2015).** *Dominance, social organisation and cooperation in the sociable weaver (Philetairus socius).* PhD Thesis, Percy FitzPatrick Institute of African Ornithology, Department of Biological Sciences, University of Cape Town, Cape Town, SA.

**Rat, M., van Dijk, R. E., Covas, R., & Doutrelant, C. (2015).** Dominance hierarchies and associated signalling in a cooperative passerine. *Behavioral Ecology and Sociobiology*, *69*(3), 437–448.

**Récapet, C., Zahariev, A., Blanc, S., Arrivé, M., Criscuolo, F., Bize, P., & Doligez, B. (2016).** Differences in the oxidative balance of dispersing and non-dispersing individuals: an experimental approach in a passerine bird. *BMC Evolutionary Biology*, *16*(1), 125.

**Rey, B., Dégletagne, C., Bodennec, J., Monternier, P.-A., Mortz, M., Roussel, D., … Duchamp, C. (2016).** Hormetic response triggers multifaceted anti-oxidant strategies in immature king penguins (Aptenodytes patagonicus). *Free Radical Biology & Medicine*, *97*, 577–587.

**Röseler, P.-F., Röseler, I., Strambi, A., & Augier, R. (1984).** Influence of insect hormones on the establishment of dominance hierarchies among foundresses of the paper wasp, Polistes gallicus. *Behavioral Ecology and Sociobiology*, *15*(2), 133–142.

**Roulin, A. (2015).** Condition-dependence, pleiotropy and the handicap principle of sexual selection in melanin-based colouration. *Biological Reviews*.

**Salin, K., Auer, S. K., Rudolf, A. M., Anderson, G. J., Cairns, A. G., Mullen, W., … Metcalfe, N. B. (2015).** Individuals with higher metabolic rates have lower levels of reactive oxygen species in vivo. *Biology Letters*, *11*(9), 20150538.

**Sapolsky, R. M. (2005).** The influence of social hierarchy on primate health. *Science*, *308*(2005), 648–652.

**Seet, R. C. S., Lee, C.-Y. J., Loke, W. M., Huang, S. H., Huang, H., Looi, W. F., … Halliwell, B. (2011).** Biomarkers of oxidative damage in cigarette smokers: Which biomarkers might reflect acute versus chronic oxidative stress? *Free Radical Biology and Medicine*, *50*(12), 1787–1793.

**Senar, J. C. (1999).** Plumage coloration as a signal of social status. In: Adams N, Slotow R (eds) Proceedings of the international ornithological congress, *Birdlife South Africa*, 22, 1669–1686

**Senar, J.C. (2006).** Color displays as intrasexual signals of aggression and dominance. *Bird Coloration*, *2*, 87–136.

**Speakman, J. R., Blount, J. D., Bronikowski, A. M., Buffenstein, R., Isaksson, C., Kirkwood, T. B. L., … Selman, C. (2015).** Oxidative stress and life histories: Unresolved issues and current needs. *Ecology and Evolution*, *5*(24), 5745–5757.

**Stephens, P. A., Buskirk, S. W., & del Rio, C. M. (2007).** Inference in ecology and evolution. *Trends in Ecology & Evolution*, *22*(4), 192–197.

**Symonds, M. R. E., & Moussalli, A. (2011).** A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology*, *65*(1), 13–21.

**Thompson, C. W., & Moore, M. C. (1991).** Throat colour reliably signals status in male tree lizards, Urosaurus ornatus. *Animal Behaviour*, *42*(5), 745–753.

**Travers, M., Clinchy, M., Zanette, L., Boonstra, R., & Williams, T. D. (2010).** Indirect predator effects on clutch size and the cost of egg production. *Ecology Letters*, *13*(8), 980-988.

**Van de Crommenacker, J., Komdeur, J., Burke, T., & Richardson, D. S. (2011).** Spatio-temporal variation in territory quality and oxidative status: A natural experiment in the Seychelles warbler (Acrocephalus sechellensis). *Journal of Animal Ecology*, *80*(3), 668–680.

**Van de Crommenacker, J., Komdeur, J., & Richardson, D. S. (2011).** Assessing the cost of helping: The roles of body condition and oxidative balance in the seychelles warbler (Acrocephalus sechellensis). *PLoS ONE*, *6*(10), e26423.

**Van Schaik, C. P. (1983).** Why Are Diurnal Primates Living in Groups? *Behaviour*, *87*(1), 120–144.

**Vitousek, M. N., Stewart, R. A., & Safran, R. J. (2013).** Female plumage colour influences seasonal oxidative damage and testosterone profiles in a songbird. *Biology letters*, 9(5), 20130539.

**Vogel, E. R. (2005).** Rank differences in energy intake rates in white-faced capuchin monkeys, Cebus capucinus: the effects of contest competition. *Behavioral Ecology and Sociobiology*, *58*(4), 333–344.

**Wegmann, M., Voegeli, B., & Richner, H. (2015).** Oxidative status and reproductive effort of great tits in a handicapping experiment. *Behavioral Ecology*, *26*, 747–754.

**White, P. A. (2007).** Costs and strategies of communal den use vary by rank for spotted hyaenas, Crocuta crocuta. *Animal Behaviour*, *73*(1), 149–156.

**Whittingham, M. J., Stephens, P. A., Bradbury, R. B., & Freckleton, R. P. (2006).** Why do we still use stepwise modelling in ecology and behaviour? *Journal of Animal Ecology*, *75*(5), 1182–1189.

**Wingfield, J. C., Ball, G. F., Dufty, A. M., Hegner, R. E., & Ramenofsky, M. (1987).** Testosterone and aggression in birds. *American Scientist*, *75*(6), 602–608.

**Wrangham, R. W. (1980).** An Ecological Model of Female-Bonded Primate Groups. *Behaviour*, *75*(3), 262–300.

**Zahavi, A., Zahavi, A., Ely, N., Ely, M. P., & Balaban, A. (1999).** *The Handicap Principle: A Missing Piece of Darwin's Puzzle*. Oxford University Press.

**Zuur, A. F., Ieno, E. N., Walker, N., Saveliev, A. A., & Smith, G. M. (2009).** Mixed effects models and extensions in ecology with R. *Statistics for Biology and Health*, 3, 57.

# Supplementary material

## Model selection procedures and model averaging

Having in account the current literature dilemma on model selection analysis (Stephens *et al.* 2007), I also followed a model averaging approach by acknowledging model selection uncertainty that is present in most of my analysis. The main difference between this method and the classical null hypothesis significant testing is that, rather than having one absolute model to explain the hypothesis, this approach takes in account several statistically indistinguishable and competing models of similar good fit when compared with corrected Akaike information criterion (AICc), which differ less than 2 AICs from the best one, by averaging them (Whittingham *et al*., 2006). Under this approach there are no p-values, instead inference is done by averaging the estimates of the all variables present in the best models. All variables are standardized to have a mean of 0 and a standard deviation of 1, so that the averages estimates are comparable. AICc values can also be used to estimate a probability of a given model to be the best approximating model, through the calculation of the Akaike weights (Symonds & Moussalli, 2011). Using these Akaike weights the relative importance of each parameter is then computed to assess the probability of a predictor to be in the true best model compared with the others (Symonds & Moussalli, 2011). This probability ranges from 0 to 1, where 1 means that the predictor is present in all selected models. Additionally, the averaged confidence intervals of each averaged estimate can be also computed and if exclude 0 can be consider as significant, similar to the classical null hypothesis testing approach (Miller 2011).

For my analysis I tested all possible combination of explanatory variables (sex, age, mass, tarsus and colony size, as well for the three-way interaction with sex and age) for each of my dependent variables: reactive oxygen metabolites (ROMs), non-specific antioxidant defences (OXY), ratio ROM/OXY, bib size and dominance score. I compared all possible combination models with mass and tarsus always included in the same model and never separated to account for body condition. Time of sampling, given its relevance in both markers variation, was also kept in all models concerning oxidative status. Colony identity was included as random effect to account for pseudoreplication. This tests were performed using the function “dredge”, to assess all possible model combinations, and the “modelavg” to assess model estimates within my range, included in the “MuMin” R package (Barton, 2013). All predictors are rescaled to a mean of 0 and a SD of 1 so that all averaged estimates are comparable.

# Model averaging results

## Oxidative status and dominance

Regarding ROMs levels, the interaction dominance*sex was present in 3 of best models (Table S1) and although with a low relative importance (0.27, Table S2), it had the highest averaged estimate of all variables and the 95% confidence intervals excluded 0 (averaged estimate= -0.614; L95%CI=-4.286, U95%=-0.528, Table S2). This result indicates that ROMs levels are significantly and positively related with dominance but only in females. Mass and tarsus were also present in 5 of the best models with a relative importance of 56, but only and the CI of Tarsus excludes 0 (averaged estimate=-0.043±0.086; L95%CI=-0.280, U95%=0.127 for Mass; averaged estimate=-0.043±0.086; L95%CI=0.024, U95%=0.422 for Tarsus, Table S2), suggesting that ROMs are not related with body condition. ROMs levels were also positively related with sampling time (averaged estimate=0.291 ± 0.103, L95%CI=0.087, U95%=0.495, Table S2).

OXY levels were not related with social rank as dominance was not present in the any of the best models (Table S1). OXY was instead related with sex (relative importance=0.80, averaged estimate=0.300 ± 0.220, L95%CI=0.019, U95%=0.732, Table S2), with higher levels of OXY in males. As with ROMs, sampling time was related with OXY levels variation (0.267 ± 0.090, L95%CI=0.088, U95%=0.446, Table S3).

The ratio between ROMs and OXY levels, in parallel to the results obtained for ROMs variation, was also related with the interaction dominance*sex (relative importance=0.53, averaged estimate=-1.373 ± 1.413, L95%CI=-4.402, U95%=-0.414). Body condition was present in 7 models but, again, only the CI of Tarsus excludes 0 (Mass: relative importance=0.53, estimate=-0.007 ± 0.077, L95%CI=-0.223, U95%=0.196; Tarsus: relative importance=0.53, estimate=0.111±0.129, L95%CI =0.001, U95%=0.416).

The other variables either had a lower relative importance and lower averaged estimates, there confidence intervals included 0, or were not present in the best models.

**Table S1**. Results from the model selection on the relation between oxidative status markers, and ratio, with individual dominance score, resuming all the models and respective AICc weights within the ΔAICc≤2 range are listed.

| | Predictors | AICc | ΔAICc | AICc weights |
|---|---|---|---|---|
| ***ROMs*** | Age[1] + ST[2] | 293.74 | 0 | 0.16 |
| | Mass + Tarsus + ST[2] | 293.81 | 0.06 | 0.16 |
| | Age[1] + Mass + Tarsus + ST[2] | 293.87 | 0.13 | 0.15 |
| | ST[2] | 293.95 | 0.2 | 0.15 |
| | DSs[3] + Age[1] + Sex + Mass + Tarsus + DSs[3]*Sex + ST[2] | 294.28 | 0.54 | 0.12 |
| | DSs[3] + Age[1] + Sex + DSs[3]*Sex + ST[2] | 295.3 | 1.56 | 0.07 |
| | DSs[3] + Age[1] + Sex + Mass + Tarsus + DSs[3]*Sex + Dss[3]*Age[1] + Age[1]*Sex + ST[2] | 295.42 | 1.68 | 0.07 |
| | DSs[3] + Mass + Tarsus + ST[2] | 295.66 | 1.92 | 0.06 |
| ***OXY*** | Sex + Colony size + ST[2] | 289.68 | 0 | 0.5 |
| | Sex | 290.67 | 0.99 | 0.3 |
| | Sex + Colony size + Age[1] | 291.47 | 1.8 | 0.2 |
| ***Ratio*** | DSs[3] + Sex + Mass + Tarsus + DSs[3]*Sex + ST[2] | 300.34 | 0 | 0.13 |
| | ST[2] | 300.47 | 0.13 | 0.12 |
| | DSs[3] + Sex + Age[1] + Mass + Tarsus + DSs[3]*Sex + ST[2] | 300.6 | 0.27 | 0.11 |
| | Mass + Tarsus + ST[2] | 301.01 | 0.68 | 0.09 |
| | DSs[3] + Sex + Age[1] + DSs[3]*Sex + ST[2] | 301.2 | 0.86 | 0.08 |
| | DSs[3] + Sex + DSs[3]*Sex + ST[2] | 301.23 | 0.89 | 0.08 |
| | Age + ST[2] | 301.44 | 1.10 | 0.07 |
| | DSs[3] + Sex + Age[1] + DSs[3]*Sex + Age[1]*Sex + ST[2] | 301.89 | 1.55 | 0.06 |
| | DSs[3] + Sex + Age[1] + Mass + Tarsus + DSs[3]*Sex + DSs[3]*Age[1] + Age[1]*Sex + ST[2] | 301.96 | 1.62 | 0.06 |
| | DSs[3] + Sex + Age[1] + Mass + Tarsus + DSs[3]*Sex + Age[1]*Sex + ST[2] | 301.98 | 1.64 | 0.06 |
| | Sex + ST[2] | 302.05 | 1.71 | 0.05 |
| | Age[1] + Mass + Tarsus + ST[2] | 302.31 | 1.97 | 0.05 |
| | Sex + Mass + Tarsus + ST[2] | 302.31 | 1.98 | 0.05 |

Reference categories: [1.] minimum age in years [2.] sampling time, [3.] standardized David's score.

**Table S2**. Summary of the relative importance and associated averaged estimates, standard errors and confidence intervals of the variables present in the best models set for the model selection analyses on the relation between oxidative status markers, and ratio, with individual dominance score.

| | Predictors | Relative Importance | Averaged Estimate (SE) | CI 95% | |
|---|---|---|---|---|---|
| ***ROMs*** | ST[1] | 1.00 | 0.291 (0.103) | **0.087** | **0.495** |
| | Age[2] | 0.58 | 0.116 (0.143) | -0.067 | 0.468 |
| | Mass | 0.56 | -0.043 (0.086) | -0.280 | 0.127 |
| | Tarsus | 0.56 | 0.125 (0.134) | **0.024** | **0.422** |
| | DSs[3] | 0.39 | 0.471 (0.831) | -0.626 | 3.061 |
| | Sex[4] | 0.27 | 0.254 (0.48) | **0.078** | **1.832** |
| | DSs[3] * Sex[4] | 0.27 | -0.641 (1.171) | **-4.286** | **-0.528** |
| | DSs[3] * Age[2] | 0.07 | -0.065 (0.268) | -1.880 | 0.019 |
| | Age[2] * Sex[4] | 0.07 | 0.037 (0.156) | -0.040 | 1.106 |
| ***OXY*** | ST[1] | 1.00 | 0.267 (0.090) | **0.088** | **0.446** |
| | Colony size | 0.70 | 0.149 (0.134) | -0.003 | 0.430 |
| | Sex[4] | 0.80 | 0.300 (0.220) | **0.019** | **0.732** |
| ***Ratio*** | ST[1] | 1.00 | 0.194 (0.103) | -0.010 | 0.398 |
| | Sex[4] | 0.67 | 0.337 (0.492) | -0.539 | 1.543 |
| | DSs[3] | 0.57 | 1.138 (1.149) | **0.451** | **3.538** |
| | DSs[3] * Sex[4] | 0.57 | -1.373 (1.413) | **-4.402** | **-0.414** |
| | Mass | 0.53 | -0.007 (0.077) | -0.223 | 0.196 |
| | Tarsus | 0.53 | 0.111 (0.129) | **0.001** | **0.416** |
| | Age[2] | 0.48 | 0.063 (0.116) | -0.146 | 0.405 |
| | Age[2] * Sex[4] | 0.17 | 0.06 (0.177) | -0.212 | 0.918 |
| | DSs[3] * Age[2] | 0.06 | -0.045 (0.217) | -1.775 | 0.183 |

Reference categories [1.] sampling time, : [2.] minimum age in years , [3.] standardized David's score, [4.] male.

## Oxidative status and bib size

Although present in some of the best models (Table S3), bib size had a low relative importance both in relation with ROMs and OXY (0.27 and 0.14; respectively; Table S4), the estimates were low (-0.028 ± 0.069; -0.005 ± 0.039; respectively; Table S4) compared to other variables present in the best models and confidence intervals always included 0. Regarding the ratio between ROMs and OXY bib size was not even present in any of the best models (table S3). Therefore there is little support for an effect of bib size on the ROMs and OXY levels, as well as the ratio between ROMs and OXY. For ratio in the analysis the CI for age excluded 0 (relative importance=0.21, averaged estimate=0.095±0.108, L95%CI =0.075 U95%CI=0.493; Table S4), indicating an effect of age on the ratio between the two markers. The other variables, including the quadratic term of bib size, either had a lower relative importance and lower averaged estimates, there confidence intervals included 0, or were not present in the best models.

**Table S3**. Results from the model selection on the relation between oxidative status markers, and ratio, with individual bib size, resuming all the models and respective AICc weights within the ΔAICc≤2 range are listed. Both linear and quadratic relations with bib size were tested, however only the linear term was present in the best models.

| | Predictors | AICc | ΔAICc | AICc weights |
|---|---|---|---|---|
| ***ROMs*** | ST[1] | 238.09 | 0 | 0.29 |
| | Age[2] + ST[1] | 238.7 | 0.61 | 0.21 |
| | Bib size + ST[1] | 239.43 | 1.35 | 0.15 |
| | Age + Bib size + ST[1] | 239.68 | 1.59 | 0.13 |
| | Mass + Tarsus + ST[1] | 239.88 | 1.79 | 0.12 |
| ***OXY*** | Sex + ST[1] | 241.85 | 0 | 0.43 |
| | Sex + Colony size + ST[1] | 242.67 | 0.82 | 0.29 |
| | Sex + Bib size + ST[1] | 244.05 | 2.2 | 0.14 |
| | Sex + Age + ST[1] | 244.06 | 2.21 | 0.14 |
| ***Ratio*** | ST[1] | 247.84 | 0 | 0.44 |
| | Age + ST[1] | 249.31 | 1.47 | 0.21 |
| | Sex + ST[1] | 249.66 | 1.82 | 0.18 |
| | Mass + Tarsus + ST[1] | 249.78 | 1.94 | 0.17 |

Reference categories: [1.] sampling time, [2.] minimum age in years.

**Table S4**. Summary of the relative importance and associated averaged estimates, standard errors and averaged confidence intervals of the variables present in the best models set for the model selection analyses on the relation between oxidative status markers, and ratio, with individual bib size.

| | Predictors | Relative Importance | Averaged Estimate (SD) | CI 95% | |
|---|---|---|---|---|---|
| ***ROMs*** | ST[1] | 1.00 | 0.415 (0.101) | **0.214** | **0.615** |
| | Age[2] | 0.34 | 0.047 (0.088) | -0.064 | 0.340 |
| | Bib size | 0.27 | -0.028 (0.069) | -0.297 | 0.090 |
| | Mass | 0.12 | -0.01 (0.045) | -0.296 | 0.130 |
| | Tarsus | 0.12 | 0.021 (0.069) | -0.032 | 0.394 |
| | Sex[3] | 0.11 | 0.013 (0.076) | -0.275 | 0.514 |
| ***OXY*** | ST[1] | 1.00 | 0.276 (0.099) | **0.080** | **0.472** |
| | Sex[3] | 1.00 | 0.47 (0.198) | **0.077** | **0.864** |
| | Colony size | 0.29 | 0.045 (0.098) | -0.089 | 0.406 |
| | Bib size | 0.14 | -0.005 (0.039) | -0.232 | 0.166 |
| | Age[2] | 0.14 | 0.005 (0.04) | -0.171 | 0.237 |
| ***Ratio*** | ST[1] | 1.00 | 0.284 (0.105) | -0.262 | 0.215 |
| | Age[2] | 0.21 | 0.095 (0.108) | **0.075** | **0.493** |
| | Sex[3] | 0.18 | -0.139 (0.21) | -0.119 | 0.310 |
| | Mass | 0.17 | 0.004 (0.113) | -0.557 | 0.279 |
| | Tarsus | 0.17 | 0.163 (0.113) | -0.222 | 0.230 |

Reference categories: [1.] sampling time, [2.] minimum age in years , [3.] male.

## Bib size correlates

Regarding the correlates of bib size, only body condition had a significant effect with mass and tarsus present in all the best models (Table S5) and with the CI of mass excluding 0 (Mass: relative importance=1, estimate=0.309 ± 0.109, L95%CI=0.092, U95%CI=0.526; Tarsus: relative importance=1, estimate=-0.034 ± 0.109, L95%CI=-0.250, U95%CI=0.182; Table S6). The other variables either had a lower relative importance and lower averaged estimates, there confidence intervals included 0, or were not present in the best models.

**Table S5**. Results from the model selection on the correlates of individual bib size, resuming all the models and respective AICc weights within the ΔAICc≤2 range are listed.

| | Predictors | AICc | ΔAICc | AICc weights |
|---|---|---|---|---|
| ***Bib size*** | Age[1] + Mass + Tarsus | 262.37 | 0 | 0.35 |
| | Mass + Tarsus | 263.39 | 1.02 | 0.21 |
| | Age[1] + Mass + Tarsus + Colony size | 263.74 | 1.37 | 0.18 |
| | Mass + Tarsus + Colony size | 264.29 | 1.92 | 0.13 |
| | Age[1] + Mass + Tarsus + Sex | 264.37 | 2.00 | 0.13 |

Reference categories: [1.] minimum age in years.

**Table S6**. Summary of the relative importance and associated averaged estimates, standard error and averaged confidence intervals of the variables present in the best models set for the model selection analyses on the correlates of individual bib size.

| | Predictors | Relative Importance | Averaged Estimate (SE) | CI 95% | |
|---|---|---|---|---|---|
| ***Bib size*** | Mass | 1.00 | 0.309 (0.109) | **0.092** | **0.526** |
| | Tarsus | 1.00 | -0.034 (0.109) | -0.250 | 0.182 |
| | Age[1] | 0.66 | 0.113 (0.112) | -0.016 | 0.361 |
| | Colony size | 0.31 | -0.043 (0.096) | -0.394 | 0.114 |
| | Sex[2] | 0.13 | -0.014 (0.079) | -0.496 | 0.271 |

Reference categories: [1.] minimum age in years, [2.] male.

## Dominance and bib size

Bib size is present in the best two models (both alone and in the interaction with dominance score; Table S7). Even though the CI included 0 (Table S8), a closer inspection to the data by testing males and females seperatly, shows that bib size is related with dominance in females (relative importance=0.67, L95%CI=0.001, U95%CL=0.127; Table S8). Nevertheless the estimate is low (estimate=0.043 ± 0.040; Table S8) and thus the effect of bib size on dominance might be weak. Age had a significant effect on males (relative importance=1.00, averaged estimate=0.148 ± 0.024, L95%CI=0.100, U95%CL=0.197; Table S8), but not on females (age was not present in the best models). The other variables either had a lower relative importance and lower averaged estimates, there confidence intervals included 0, or were not present in the best models.

**Table S7**. Results from the model selection on the linear relation between dominance score and bib size, resuming all the models and respective AICc weights within the ΔAICc≤2 range are listed. The analysis was performed first for all individuals, then separately for each sex.

| ***Dominance score*** | **Predictors** | **AICc** | **ΔAICc** | **AICc weights** |
|---|---|---|---|---|
| *All individuals* | Age[1] + Sex + Age[1]*Sex + Bib size | -43.63 | 0 | 0.46 |
| | Age[1] + Sex + Age[1]*Sex + Bib size + Bib size*Sex | -42.6 | 1.04 | 0.27 |
| | Age[1] + Sex + Age[1]*Sex | -42.54 | 1.10 | 0.27 |
| *Only males* | Age[1] | -26.64 | 0 | 0.7 |
| | Age[1] + Bib size | -24.97 | 1.67 | 0.3 |
| *Only females* | Bib size | -14.87 | 0 | 0.67 |
| | Null | -13.43 | 1.44 | 0.23 |

Reference categories: [1.] minimum age in years.

**Table S8**. Summary of the relative importance and associated averaged estimates, standard errors and averaged confidence intervals of the variables present in the best models set for the model selection analyses on the linear relation between dominance and individual bib size.

| ***Dominance score*** | **Predictors** | **Relative Importance** | **Averaged Estimate (SD)** | **CI 95%** | |
|---|---|---|---|---|---|
| *All individuals* | Age[1] | 1.00 | 0.278 (0.029) | -0.056 | 0.058 |
| | Sex[2] | 1.00 | 0.001 (0.029) | **0.235** | **0.383** |
| | Age[1]: Sex[2] | 1.00 | 0.309 (0.037) | **0.071** | **0.220** |
| | Bib size | 0.73 | 0.145 (0.037) | -0.010 | 0.103 |
| | Bib size * Sex[2] | 0.27 | 0.046 (0.029) | -0.124 | 0.032 |
| *Only males* | Age[1] | 1.00 | 0.148 (0.024) | **0.100** | **0.197** |
| | Bib size | 0.30 | 0.035 (0.090) | -0.150 | 0.380 |
| *Only females* | Bib size | 0.67 | 0.043 (0.040) | **0.001** | **0.127** |

Reference categories: [1.] minimum age in years, [2.] male.

Although present in the best models (Table 9), quadratic term of bib size had a very low relative importance and confidence intervals included 0 (relative importance=0.20, estimate=0.108 ± 0.313, L95%CI=-0.461, U95%CL=1.552; Table S10). The other variables either had a lower relative importance and lower averaged estimates, there confidence intervals included 0, or were not present in the best models.

**Table S9**. Results from the model selection on the quadratic relation between dominance and bib size in the, resuming all the models and respective AICc weights within the ΔAICc≤2 range are listed.

| | Predictors | AICc | ΔAICc | AICc weights |
|---|---|---|---|---|
| ***Dominance score*** | Age[1] + Sex + Age[1]*Sex + Bib size | -43.63 | 0 | 0.46 |
| | Age[1] + Sex + Age[1]*Sex + Bib size + Bib size*Sex | -42.60 | 1.04 | 0.27 |
| | Age[1] + Sex + Age[1]*Sex | -42.54 | 1.10 | 0.27 |
| | Age[1] + Sex + Age[1]*Sex + Bib size + Bib size[^2] | -42.39 | 1.24 | 0.20 |

Reference categories: [1.] minimum age in years, [^2] bib size quadratic term.

**Table S10**. Summary of the relative importance and associated averaged estimates, standard errors and averaged confidence intervals of the variables present in the best models set for the model selection analyses on the quadratic relation between dominance and individual bib size.

| | Predictors | Relative Importance | Averaged Estimate (SD) | CI 95% | |
|---|---|---|---|---|---|
| ***Dominance score*** | Sex[2] | 1.00 | 0.308 (0.037) | **0.234** | **0.382** |
| | Age[1] | 1.00 | 0.001 (0.029) | -0.056 | 0.058 |
| | Age[1] * Sex[2] | 1.00 | 0.144 (0.037) | **0.070** | **0.219** |
| | Bib size | 0.79 | -0.021 (0.161) | -0.384 | 0.332 |
| | Bib size * Sex[2] | 0.22 | -0.010 (0.026) | -0.124 | 0.032 |
| | Bib size[^2] | 0.20 | 0.108 (0.313) | -0.461 | 1.552 |

Reference categories: [1.] minimum age in years, [2.] male, [^2] bib size quadratic term.

# References

**Stephens, P. A., Buskirk, S. W., & del Rio, C. M. (2007).** Inference in ecology and evolution. *Trends in Ecology & Evolution*, *22*(4), 192–197.

**Symonds, M. R. E., & Moussalli, A. (2011).** A brief guide to model selection, multimodel inference and model averaging in behavioural ecology using Akaike's information criterion. *Behavioral Ecology and Sociobiology*, *65*(1), 13–21.

**Whittingham, M. J., Stephens, P. A., Bradbury, R. B., & Freckleton, R. P. (2006).** Why do we still use stepwise modelling in ecology and behaviour? *Journal of Animal Ecology*, *75*(5), 1182–1189.